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***Corbicula fluminea* as a Bioaccumulation Indicator Species: A Case Study at the Columbia and Willamette Rivers**

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Final report

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Abstract: The freshwater Asiatic Clam, *Corbicula fluminea*, is found in abundance throughout North America. *C. fluminea* are primarily considered filter-feeders; however, they routinely bury in the sediment for extended periods and filter interstitial sediment water (pore water) or pedal-feed. *C. fluminea* shows promise as a model trophic-niche freshwater test organism or as an indicator species for bioaccumulation studies for the assessment of contaminants in sediments as part of dredging, restoration, remediation, and monitoring evaluations. In August and September 2005, 32 nearshore locations were sampled for *C. fluminea* along the Columbia River from Vista Park near Skamokawa, Washington (River Mile 32) to Warrendale, Oregon (River Mile 147). Four additional samples were collected in the lower Willamette River, near its confluence with the Columbia River (Columbia River Mile 102). Tissue samples were analyzed for semi-volatile compounds (including polycyclic aromatic hydrocarbons, PAH); chlorinated pesticides; polychlorinated biphenyl (PCB Aroclors and 209 congeners); polybrominated diphenyl ethers (PBDE; fire retardants); organotins; and four metals (Hg, Pb, Zn, Cd). All clam tissue had detectable levels of many of the chemicals analyzed. Statistical relationships among sampling stations were elucidated using exploratory multivariate statistical techniques. Relative abundances of major constituents were superimposed on regional maps displaying the sampling stations. A mid-reach point source for PCBs was identified, as were localized areas of DDTs, PBDEs, and PAHs.

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Preface

One of the more significant challenges for the assessment of sediment-associated contaminants is the high level of uncertainty in the assessment of bioavailability. Because of the spatial and temporal variability of sediments and their associated contaminants, organisms may be exposed and accumulate different levels of chemicals in their tissues. The current report addresses these challenges by demonstrating the use of field collected freshwater clams, *Corbicula fluminea*, as an indicator of contaminants that may be bioavailable and bioaccumulate in a food web. The study focuses on more than 100 miles of the Columbia River in Washington and Portland, Oregon USA. Advanced statistical procedures were used to cluster samples based on their location and contaminants present in tissue.

This research was conducted through a joint effort with the Portland District, U.S. Army Corps of Engineers (USACE) and the U.S. Army Engineer Research and Development Center (ERDC), Environmental Laboratory (EL). This work was conducted under the general supervision of Dr. Robert P. Jones, Acting Chief, and Mr. Warren Lorentz, Chief, Environmental Risk Assessment Branch, EL and Dr. Richard E. Price, Chief, Environmental Processes and Engineering Division, EL. At the time of publication of this report, Dr. Beth Fleming was Director of the Environmental Laboratory.

COL Gary E. Johnston was Commander and Executive Director of ERDC. Dr. James R. Houston was Director.

Unit Conversion Factors

Multiply	By	To Obtain
cubic feet	0.02831685	cubic meters
feet	0.3048	meters
inches	0.0254	meters
miles (nautical)	1,852	meters
square miles	2.589998 E+06	square meters

1 Introduction

Assessing the extent and fate of sediment-associated contaminants in large study areas, such as the estuarine and riverine systems of the Columbia River basin, is difficult and can be time-consuming and expensive (Culp et al. 2000, U.S. Environmental Protection Agency (USEPA) 2000). Often these studies rely on food web models that require large amounts of field-collected data as well as multiple assumptions such as physical/chemical characteristics, kinetics rates, and physiological parameters of receptor organisms (Arnot and Gobas 2004, Iannuzzi et al. 1996). Furthermore, a compilation of site-specific data is frequently limited in its ability to adequately describe conditions for the entire basin (von Stackelberg et al. 2002). Sites are often characterized using traditional tools such as sediment concentrations and tissue chemistry from benthic invertebrate bioaccumulation tests. However, these approaches have the limitation that spatial and temporal trends are not resolved nor adequately described.

Several methods and protocols have been developed and utilized to address these data gap issues. One method uses semi-permeable membrane devices (SPMDs) or polypropylene sheets, which mimic the bio-membrane's ability to store contaminants (Petty et al. 2004). Another method exposes caged bivalves to sediment and/or the water column in situ. A third method is the systematic collection of various organisms (fish, bivalves, insects, etc.) from the study area. All three methods have their strengths and limitations (Crane et al. 2007).

The high cost of determining bioaccumulation potential by using sediment/tissue testing methods or by deploying SPMDs necessitates that only a few samples will represent large areas. The membranes must be deployed for a fixed time and then be retrieved, and the information gathered only applies for the duration and location of the deployment. Caged bivalves also must be deployed and retrieved, but they have the advantage of maintaining physiological processes such as uptake rate, metabolism, and elimination. However, the SPMDs and caged bivalves do not necessarily reflect steady-state bioaccumulation that would be present in the collection of resident organisms. These methods do have advantages over field-collected animals in that exposure location can be controlled,

sensitive species may be used, and organism selection can be based on well-studied animal models.

Numerous fish samples collected throughout the Columbia River system have been shown to contain contaminants as summarized by Johnson et al. (2007) and detailed in a regional study of the Columbia River (Lower Columbia River Estuary Partnership (LCREP) 2007). However, determining contaminant source and exposure is problematic because of fish mobility. In large study areas, such as riverine systems, collection of either indigenous or non-native clams has great potential as an effective tool for monitoring bioaccumulation potential. Because clams can be collected from more sampling stations, processed more cheaply, and are more likely to be at equilibrium, they have added advantages over fish or SPMDs.

The use of bivalves for evaluating bioaccumulation of contaminants in sediments has been investigated and employed in dredged material evaluations, monitoring stormwater runoff and release, and risk assessment for remediation. The saltwater clam, *Macoma sp.*, has been a recommended marine bioaccumulation test species for whole sediment in national and regional sediment-quality testing manuals (USEPA/U.S. Army Corps of Engineers (USACE) 1991, 1998; USACE 1998). Both the zebra mussel (*Dreissena polymorpha*) and Asian clam (*Corbicula fluminea*) are promising species for freshwater bioaccumulation tissue studies (Smolders et al. 2003).

The freshwater clam *C. fluminea* is a non-native, filter-feeding clam found in abundance throughout most systems in the northwestern United States. Native to China, Korea, and southeastern Russia, the earliest verifiable record of this species in North America was at Nanaimo, Vancouver Island, British Columbia in 1924 (Counts 1981). Asian clams are believed to have established a viable population on the west coast of the United States sometime prior to 1938 (Cherry et al. 1980). Currently, *C. fluminea* are widespread and found throughout most of the United States including Hawaii (Invasive Species Specialist Group (ISSG) 2007). While they are typically considered a freshwater species, they are salt-tolerant to 13 ppt for short periods, and higher if allowed to acclimate. Estuarine populations have been documented in the San Francisco and Chesapeake Bays. However, to date their distribution in the brackish lower Columbia River estuary has not been documented in the literature.

C. fluminea is a promising species for bioaccumulation tissue studies for several reasons. Their lifespan can reach a maximum of 7 years but normally ranges from 3-5 years. Though the clams can be found in any habitat, they prefer flat areas with combinations of fine clean sand, clay, and coarse sand. Obviously, the clam has limited mobility and is a good indicator of site-specific potential for bioaccumulation. It fills the stated need for a trophic niche freshwater test organism for bioaccumulation that has sufficient tissue volume to run multiple contaminants of concern (CoC) analyses. The clam has been shown to bioaccumulate CoC such as DDT, PCBs, and PAHs (Buck 2004; Johnson and Fishman 1993; Tran et al. 2002; Phelps 2003, 2005; Versteeg and Rawlings 2003) and metals (Fournier et al. 2005; Croteau et al. 2004; Inza et al. 1998). Although *C. fluminea* are filter feeders, they routinely bury in the sediment for extended periods and use interstitial sediment water (pore water) as a filter water source, potentially making them a good indicator of sediment contamination. Additional studies have explored their use in assessing toxicity of contaminants in situ through the use of caged exposures (Hull et al. 2002, 2004).

Several studies have been conducted using *C. fluminea* as a test organism for laboratory bioaccumulation tissue analyses with Columbia River basin sediments (USACE 2002, 2004; USEPA 2007a). In 2001, USACE assessed *C. fluminea* during a study to identify reference sediment sampling sites (reference sediment is used as a control in sediment bioaccumulation laboratory tests), running both *Lumbriculus variegatus* and *C. fluminea* in a limited side-by-side bioaccumulation test species comparison. In 2003, USACE ran paired 28-day *L. variegatus* and *C. fluminea* bioaccumulation tests on dredged material from the Willamette River Federal Navigation Channel. Results of the comparison of uptake of metals were mixed, depending on the metal. For organics, lipid-normalized concentrations of PAHs and PCBs were consistently lower in the *C. fluminea* tissue; with differences as much as three times lower in the clams for the PAHs. While PCBs were detected in all *L. variegatus*, no PCBs were detected in the paired clams. Whether or not steady state was reached is still an unanswered question.

As part of the USEPA Willamette River superfund remedial investigation and feasibility study, the identified Potentially Responsible Parties (PRP) formed a study group called the Lower Willamette Group (LWG)

(USEPA 2007a). The LWG sampled clams and sediment from 33 locations in the Lower Willamette River in 2005. While the LWG analyzed in situ clams, they also conducted paired laboratory bioaccumulation tests using both *L. variegatus* and *C. fluminea*. At this time the data are still under review by the USEPA and its federal, state, and tribal partners.

In 1992-1993, Fishman Environmental Services used *C. fluminea* collected from Fitzpatrick Island (RM 31.3) in developing a long-term bio-monitoring program for the Columbia Slough near Portland, Oregon for the City of Portland Bureau of Environmental Services (Johnson and Fishman 1993). Studies in the Anacostia River estuary of Washington, D.C. (Phelps 2003, 2005) used translocated clams to assess contaminant exposure. Future Anacostia River management strategies call for concurrent deployment of *C. fluminea* clams and SPMDs.

While some limited in situ clam tissue studies have been conducted on the Columbia River, this is the first large-scale study targeting a significant stretch of the river (115 miles). In a study by Buck (2004), *C. fluminea* tissue was collected from the Julia Butler Hanson National Wildlife Reserve (RM 35) as part of the 1991 bi-state effort. The Washington Department of Ecology (DOE) collected clams from 18 locations below Dalles Dam (RM 192) and analyzed PCB uptake in response to a January 2004 transformer oil spill. Limited SPMDs also were deployed to supplement an ongoing DOE SPMD study (Johnson and Norton 2005). Other studies (Buske 2005; Kaltofen and Carpenter 2005) looked at radionuclide levels in clams along the 50-mile Hanford Reach (~RM 346-396).

The present study objectives were to:

1. Determine if *C. fluminea* bioaccumulates chemicals of concern in the Columbia River system.
2. Determine the level of utility for *C. fluminea* in the spatial assessment of contaminants in a large watershed.
3. Evaluate the role of *C. fluminea* for the assessment of contaminant bioaccumulation in the context of other lines of evidence (e.g., laboratory bioassays).

2 Materials and Methods

Study area

The Columbia River basin drains 259,000 square miles covering parts of Oregon, Washington, Idaho, and Montana as well as very small portions of California, Utah, Wyoming, and Nevada. The Columbia River itself originates in Canada's Columbia Lake, flowing 1,214 miles to the Pacific Ocean near Astoria, Oregon. The Columbia River is the largest river in volume flowing into the Pacific Ocean (http://en.wikipedia.org/wiki/Pacific_Ocean) from the Western Hemisphere (http://en.wikipedia.org/wiki/Western_Hemisphere), and is the fourth largest by volume in North America, with an annual flow averaging 262,000 ft³/sec. The present study area (Figure 1) consisted of that portion of the Columbia River from Warrendale, Oregon (RM 142) downstream of the Bonneville Dam to Skamokawa, Washington at RM 32. The river in this reach occupies a single main channel with occasional small side channels around small, low islands. Ocean tides influence water surface elevations upstream to Bonneville Dam and the Willamette Falls and can create slack water conditions. Flow reversals are dependent on river location, height of the tide, and flow condition. During low flow and high tide, current reversals can reach RM 90 on the Columbia. During high flows in the Columbia River and low flows on the Willamette River, the Columbia River can flow up the Willamette River and discharge through the Multnomah Channel at Willamette RM 2.

In August and September 2005, the low-flow period on the Columbia River, 36 nearshore locations were sampled for *C. fluminea*. The collection team consisted of one to four persons with access from shore or by boat. Clams were collected by hand or using small hand rakes. Collection using a larger clam rake had been attempted but proved cumbersome and inefficient. Sampling was scheduled for low tide to allow access to as large a shoreline area as possible. Clams were collected from the wetted shore out to a depth of about 3 ft of water. Photos from the sample sites and collection are shown in Figures 2 to 7.

Of the 36 samples collected, 7 were collected in tributaries to the Columbia River. Four samples were collected in the lower Willamette River, and one sample each was collected from the mouth of Multnomah Channel near

the town of St. Helens, Oregon; the mouth of Lake River across the river from St. Helens; and the “Old Mouth” of the Cowlitz River at Longview, Washington. While sampling stations were selected to provide a distribution of station locations along the river, more stations were sited near the Portland/Vancouver metropolitan area.

Most of the areas sampled consisted of sandy beaches. Stations with fine-grained material included Stations 10, 11, and 31 on the Willamette River, Station 28 on Lake River, and Station 30 at the Mouth of the Multnomah Channel. Station 2 at Stella, Washington, Station 6 across from Camas, Washington, and Station 7, Lady Island, had a mix of sand and gravel areas. At Stations 2, 6, and 30, the bulk of the clams collected were not buried but on the surface. Station 30 was unique in that the substrate where the clams were found and collected consisted of a consolidated clay bank. In all other locations, the clams were buried in the upper 1-2 in. of the sediment.

Clams in sufficient number were found at most locations except the beach along Rainier, Oregon; Columbia City, Oregon; and the upstream end of Reed Island, Washington. At Rainier (RM 67), clams were found but not in sufficient number to conduct analyses. At Columbia City (RM 84), the shore was mostly basalt rock with very few clams. The area upstream of Reed Island (RM 127) appeared to be suitable clam habitat but contained no evidence of clams, possibly due to high energy river flow. Clams were found approximately 1 mile downstream of Reed Island.



Figure 1. Map of sample station locations.



Figure 2. Collection of *Corbicula fluminea* near Stella, Washington (RM 56; SG-02). The picture shows coarse sand, gravel, and cobble at the collection site.



Figure 3. US Moorings, Willamette River (RM 6.2), showing fine-grained sandy silt.



Figure 4. Collection near Government Island (RM 112.5, SG-33), downstream of the I-205 bridge between Oregon and Washington, showing a clean medium sand.



Figure 5. Hand collection of clams, Government Island (RM 112.5; SG-33)



Figure 6. Typical representation of clams collected from a sample site.



Figure 7. Large clams and relative scale of size.

Chemical analysis

Following collection, the clams were placed on ice in coolers, then either frozen or shipped immediately to analytical chemistry laboratories for analysis. Whole undepurated clams were shucked and the tissue was composited from each sample location for analysis.

Tissue samples were analyzed for the following bioaccumulative constituents: 32 semi-volatile compounds (including PAH); chlorinated pesticides; 209 polychlorinated biphenyl (PCB) congeners; 7 PCB Aroclors; 11 polybrominated diphenyl ether (PBDE) congeners; organotins, and 4 inorganic metals including mercury (Hg), lead (Pb), zinc (Zn), and cadmium (Cd). The analytical methods used by the laboratories and sample identifications and dates of collection are listed in Appendix B.

Multivariate statistical methods and data interpretation

Data were analyzed by multivariate statistical techniques including non-metric Multidimensional Scaling (MDS) and Hierarchical Classification

(clustering) using Euclidean distance as the distance measure. Prior to analysis all data were first converted to the same scale (ppb) to facilitate simultaneous comparisons, and nondetects were expressed as zeros. PCB and PBDE data were normalized to lipid content. Data for all analytes were standardized by calculating their percent contribution to each sample. To reduce the disproportionate impact of the contribution of the most dominant analytes and approximate normality, data were transformed by taking the square root of each value as recommended by Clarke and Warwick (2001). During analysis of the individual contaminant classes it was necessary to apply a fourth-root transformation to the PBDE data.

MDS results, though multidimensional in nature, are displayed as two-dimensional graphs in which the axes are those dimensions that account for the largest amount of variability in the data. MDS plots were interpreted by overlaying them with the clustering results. The presence of similar groupings of samples in both clustering and MDS is considered to be a good indication of the robustness of the analyses. A bootstrapping test (SIMPROF) assisted in determining which levels from the clustering results to plot by indicating which cluster groups might have occurred by random chance alone. Details of the distribution of contaminants among stations were explored by overlaying concentrations on the joint cluster-MDS plots (Biplots) and by performing Nodal Analysis. Nodal Analysis constructs a table from the results of both Q-mode (stations) and R-mode (contaminants) cluster analyses.

Because the methods employed require that data be complete for all samples (stations) and variables (contaminant concentrations), those stations where specific contaminants were not found and those contaminants for which all values were nondetects had to be excluded from the analysis. Careful review of the data suggested that removal of incomplete data would not compromise interpretation of the data set.

Data analysis was conducted in two stages. The first step was to perform the analyses on data aggregated into major classes (e.g. Total PAH, Total PCB, etc.). In the second stage of analysis, the individual classes of contaminants were analyzed separately to examine the distribution patterns of individual chemicals.

3 Results

Analytical chemistry results for all detected and qualified analytes are reported in Appendix A. The chlorinated pesticide data were deemed unreliable due to analytical chemistry problems and were not subjected to further analysis. These rejected data are further explained in the data validation summary in Appendix B. Based on the frequency of detection, magnitude, relevance for bioaccumulation, and data validation results, PCBs, PBDEs, and PAHs were identified as the primary chemical constituents for further evaluation of spatial trends and utility of *C. fluminea* for use as an indicator for contaminant bioaccumulation. Data were analyzed using multivariate techniques as described above and shown in the joint cluster MDS plots in Appendix C. Statistical analyses for summed compound classes for PCB, PBDE, and PAH compounds are described and shown below.

Metals

Metals that were analyzed did not appear to have any specific trends and did not vary significantly across the sample locations. Using MDS-clustering biplots (Appendix C), the metals formed a singleton group (Station 31) and a large group with several subgroups. Station 31 had relatively low concentrations of zinc (19.3 µg/g), moderate levels of mercury (46.5 ng/g), and undetectable levels of cadmium (< 0.1 µg/g). Within the large group, the first subgroup was a singleton, Station 19, which had the highest mercury concentrations (171 ng/g). A second subgroup consisting of Stations 1 and 20A had moderate to high levels of lead (0.25 – 0.587 µg/g). The third subgroup, Stations 16, 23, 27, and 35, was characterized by undetectable levels of lead (< 0.1 µg/g) and moderate concentrations of mercury (21.4 - 40.2 ng/g). Subgroup 4 contained Stations 4, 8, 10, 14, 17, 18, 24, 26, and 29, and was notable for moderate concentrations of mercury (20 - 43.6 ng/g) and moderate to high levels of zinc (21.3 – 32.8 µg/g). The remaining stations had no detectable mercury, with detection limits typically below 40 ng/g.

Polychlorinated Biphenyls (PCBs)

Total PCBs (sum of all analyzed congeners) in *C. fluminea* had a median concentration of 1,050 pg/g lipid and a range of 307 to 102,000 pg/g lipid (Figure 8). The joint cluster MDS plot for PCBs had four major groups, two of which were made up exclusively of Upper Columbia stations (Figures 9 and 10). The first of these groups contained two subgroups: Stations 5, 6, 7, 8, 18, 19, and 20; and Stations 17, 32, and 34. Both subgroups had proportionally high concentrations of PCB 11 and low concentrations of most other congeners; the first subgroup also included 39 congeners that were not detected in the second subgroup. PCB 11 was a dominant contributor to the total PCBs measured in *C. fluminea*, with a median concentration of 2,200 pg/g lipid and a range from 440 to 5,100 pg/g lipid.

Stations 12, 13, 25, and 26 (Lower Columbia) formed a single group characterized by moderate to high concentrations of total lipid-normalized PCBs (3,032 to 102,000 pg/g lipid). The bulk of the Lower Columbia and all of the Willamette stations were found in a single group, which in turn contained four subgroups: 1) Stations 30 and 31, 2) Stations 35 and 36, 3) Station 27 alone, and 4) all the Willamette stations (except 31) plus the remaining Lower Columbia stations. Stations 30 and 31 were characterized by the lowest concentrations of congener 11, Stations 35 and 36 by below-average concentrations of most congeners, Station 27 by moderate concentrations of congener 11 and moderate to low concentrations of all other congeners, and the remaining stations by moderate to low concentrations of most detected congeners and high concentrations of a few. Overall, *C. fluminea* sampled above river mile 108 had a different composition and lower concentrations of PCB congeners than clams from the Lower Columbia and Willamette stations (Figure 10).

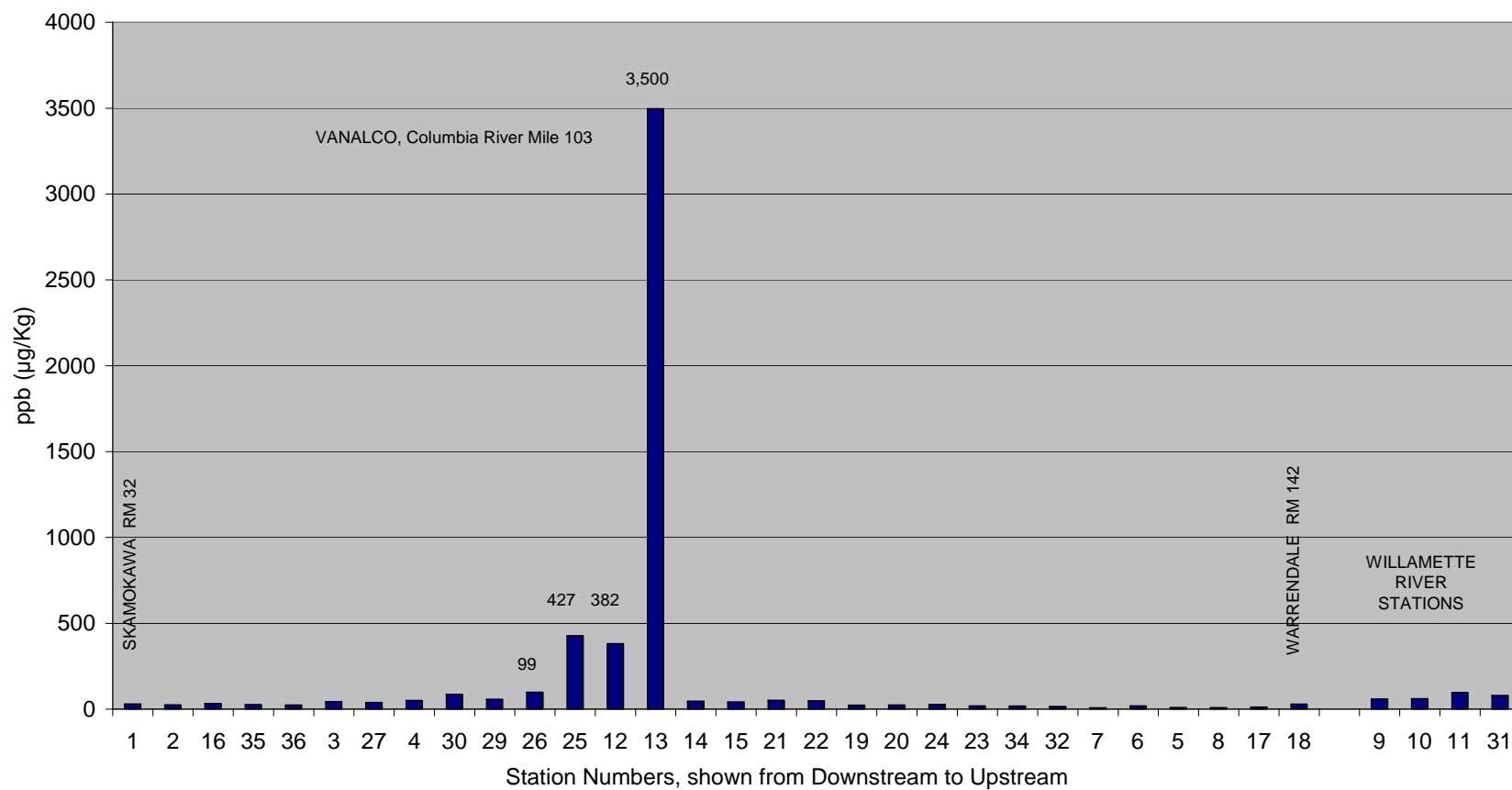


Figure 8. Total PCB (not lipid normalized) in *Corbicula fluminea* by station number in the Columbia and Willamette Rivers.

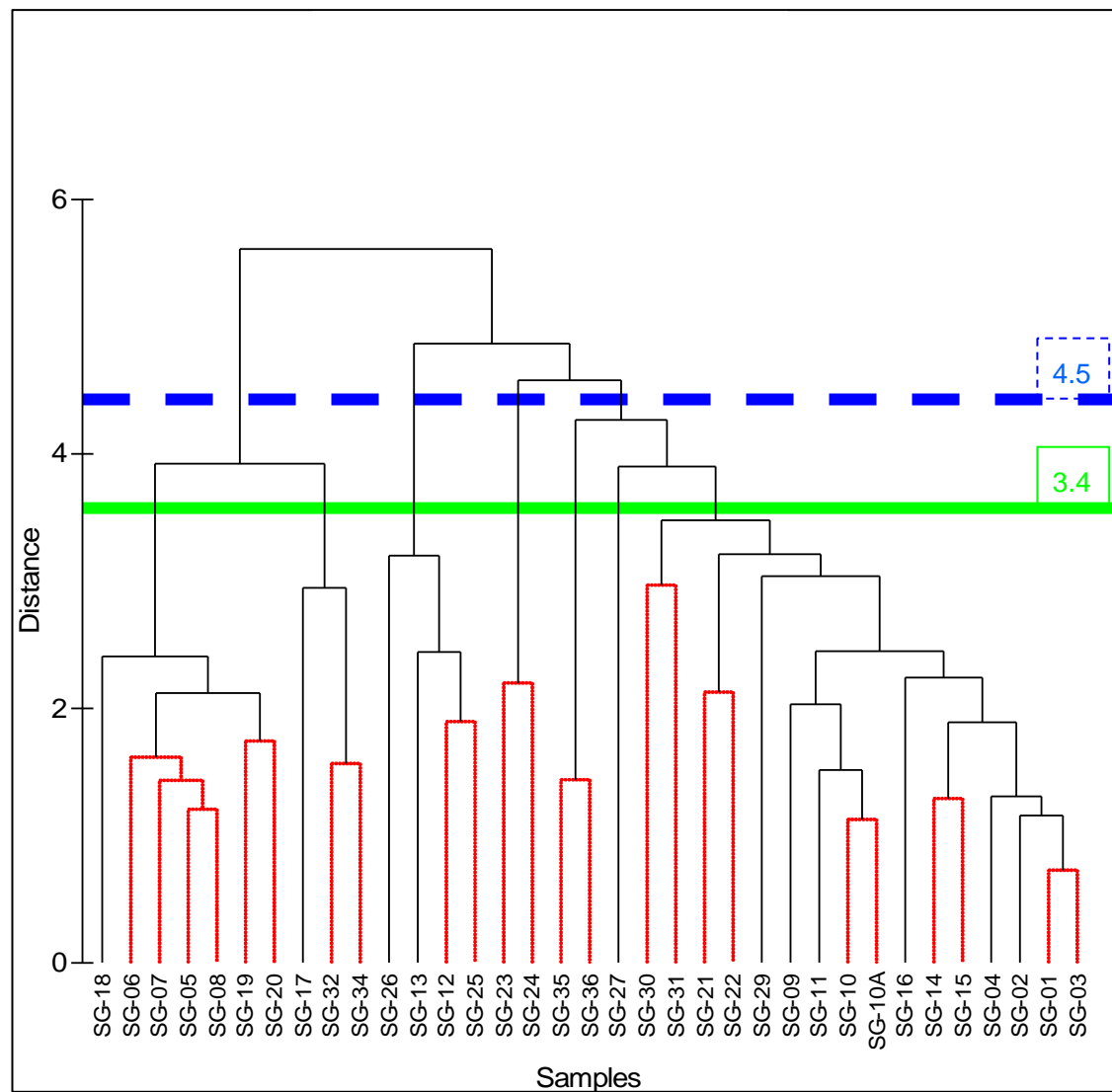


Figure 9. Hierarchical classification (clustering) of stations using lipid-normalized PCB concentrations in *Corbicula fluminea*. Groupings in the MDS-cluster plot (Figure 10) are separated by the green (3.4) and blue (4.5) Euclidean distance lines.

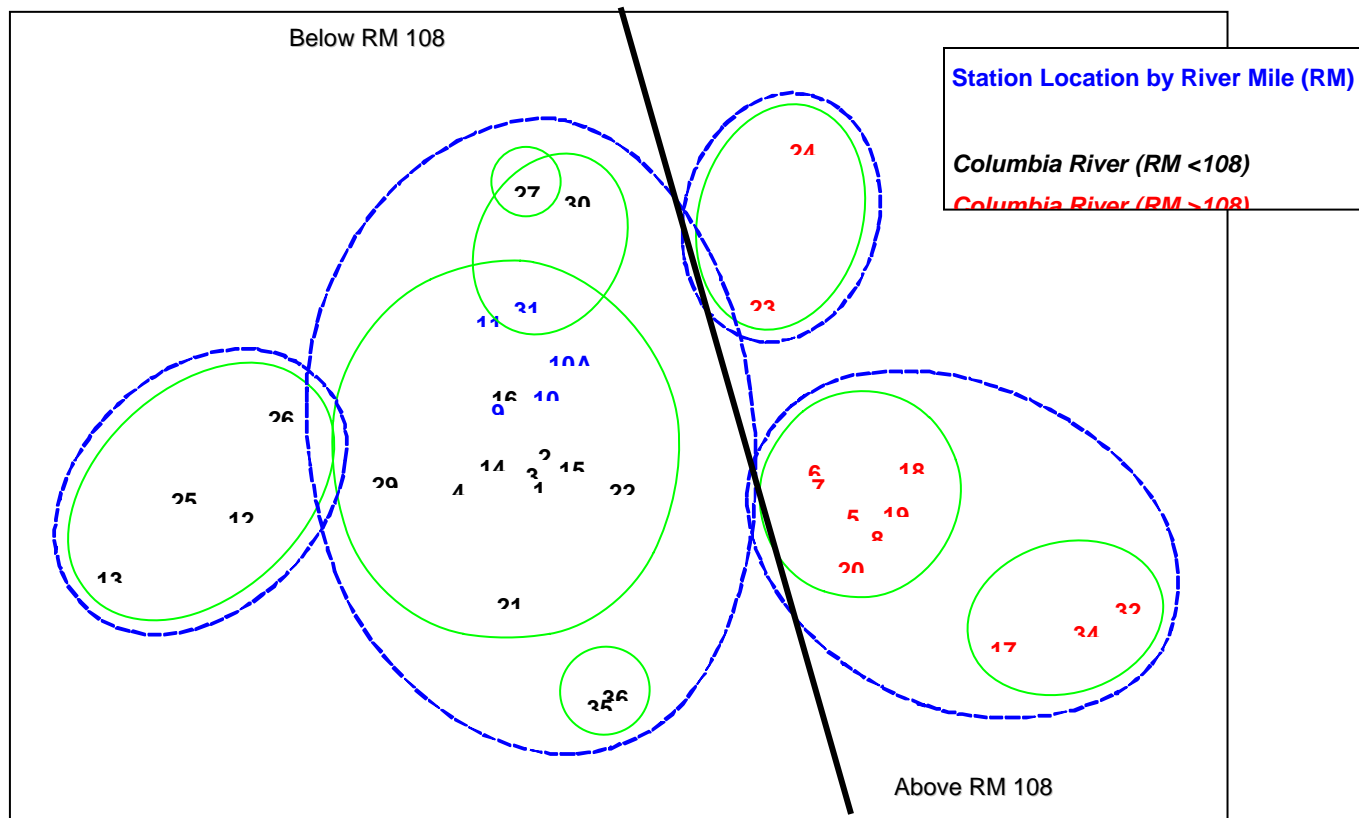


Figure 10. MDS plot for lipid-normalized total PCB concentrations in *Corbicula fluminea*. Euclidean distance is represented by green (3.4) and blue (4.5) lines.

Polybrominated Diphenyl Ether Congeners (PBDE)

Nine of 11 PBDE congeners were detected in *C. fluminea* tissues. The sum of PBDE congeners in *C. fluminea* had a median concentration of 268 ng/g lipid and ranged from 5.4 to 6,907 ng/g lipid. These lipid-normalized PBDEs also displayed the same geographic pattern as the PCB data with Upper Columbia stations generally having the lowest concentrations (Figure 11). The data formed three groups (Figures 12 and 13): Station 8 alone; Stations 6, 7 and 24; and all remaining stations roughly grouped together into two subgroups. One subgroup contained Stations 1 through 5, 9, 11 through 20, 25, 32, and 34 through 36, most of them in very close proximity in terms of statistical similarity. The other subgroup consisted of Stations 10, 10A, 21, 23, 26, 27, 29, and 30. Station 8 had low but detectable concentrations of PBDE congeners 28, 66, and 154 only. Stations 6, 7, and 24 also had low but detectable concentrations of these three congeners plus low concentrations of PBDE 153 and 100. The first subgroup of the remaining stations (Stations 1 to 5, etc.) had low to moderate concentrations of all congeners except PBDE 99 and 183 while the other subgroup (Stations 10, 10A, etc.) generally had moderate concentrations of all congeners except PBDE 183. All PBDE congeners were found at Stations 29 and 30, in concentrations that were at least an order of magnitude higher than most other stations. The distribution of non-lipid normalized PBDE congeners is shown in Table 1 and includes selected congeners from all sample locations. The dominant PBDE congener in this system is PBDE 47 followed by PBDEs 100 and 99, respectively.

Table 1. Distribution of PBDE congeners in tissues of *C. fluminea*.

Congener	Mean pg/g	% of Total
PBDE 28	256	1
PBDE 47	10,975	58
PBDE 66	443	2
PBDE 85	374	2
PBDE 99	1,839	10
PBDE 100	4,258	22
PBDE 138	nd	<1
PBDE 153	445	2
PBDE 154	390	2
PBDE 183	nd	<1
PBDE 209	nd	<1
Total	18,983	100

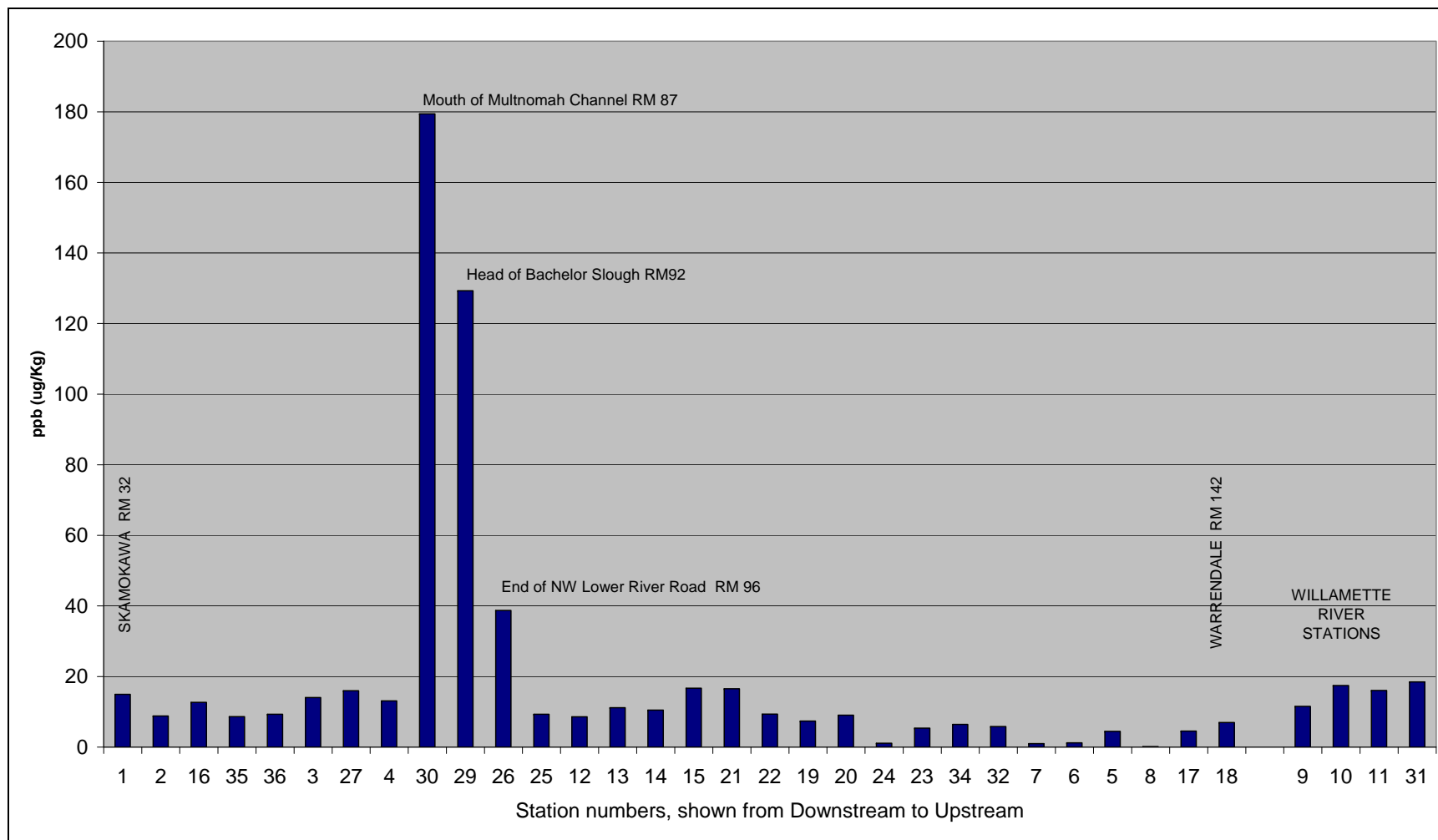


Figure 11. Total PBDE in *Corbicula fluminea* by station number in the Columbia and Willamette Rivers.

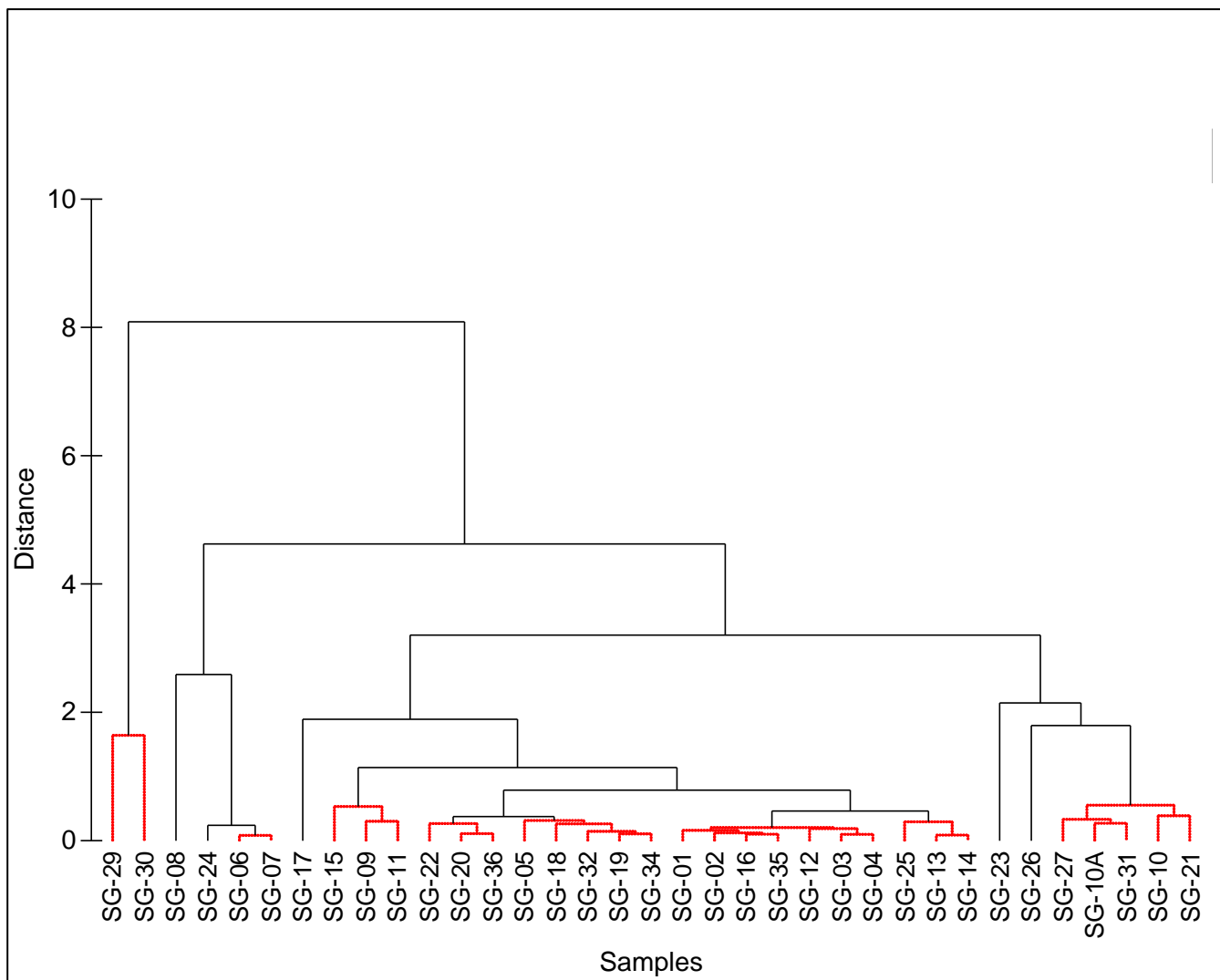


Figure 12. Hierarchical classification of lipid normalized total PBDE concentrations in *Corbicula fluminea*.

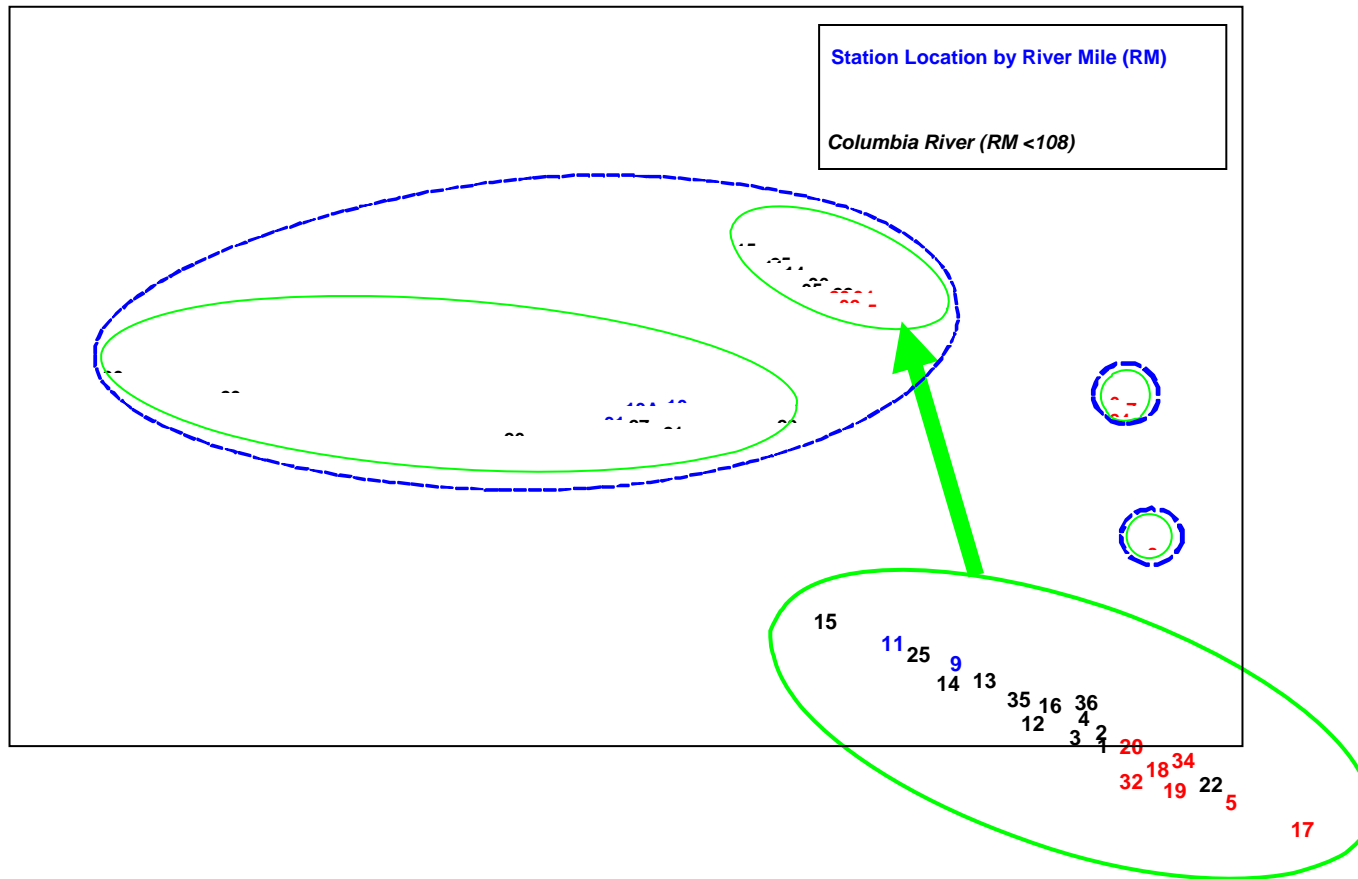


Figure 13. MDS plot for lipid-normalized total PBDE concentrations in *Corbicula fluminea*. Euclidean distance is represented by green (3.2) and blue (4.9) lines.

Polycyclic Aromatic Hydrocarbon Compounds (PAHs)

PAHs displayed the same geographic pattern as the PCB and PBDE data, with Upper Columbia stations generally having the lowest concentrations of PAHs (Figure 14). PAHs measured in tissues of *C. fluminea* were not lipid normalized. The concentration of total PAH had a median value of 47.6 ng/g and ranged from 20.4 to 4,692 ng/g. In the PAH MDS-cluster plot, stations were classified into six groups: 1) Station 7 alone; 2) Stations 8, 17, and 18; 3) Stations 9, 15, and 22; 4) Stations 11, 17, 31, 35, and 36; 5) Station 30 alone; and 6) all remaining stations (Figures 15 and 16). Station 7 was unique in that it had the highest concentration of anthracene, but no detectable acenaphthylene, dibenz[a,h]anthracene, indeno[1,2,3-cd]pyrene, benzo[ghi]perylene, benzo[b]fluoranthene or benzo[k]fluoranthene, and only low levels of all other PAHs. Stations 8, 17, and 18 also lacked dibenz[a,h]anthracene, indeno[1,2,3-cd]pyrene, benzo[ghi]perylene, benzo[b]fluoranthene and benzo[k]fluoranthene, and had low to moderate levels of the remaining compounds. Stations 9, 15, and 22 had mostly moderate concentrations of nearly all the PAHs, while Station 30 had low to moderate concentrations of all except acenaphthylene and dibenz[a,h]anthracene. Stations 11, 17, 31, 35, and 36 had moderate to high concentrations of most of the PAHs. Station 31, in particular, had the highest concentrations of all the PAHs except anthracene. The remaining stations all had low to moderate concentrations of most PAHs.

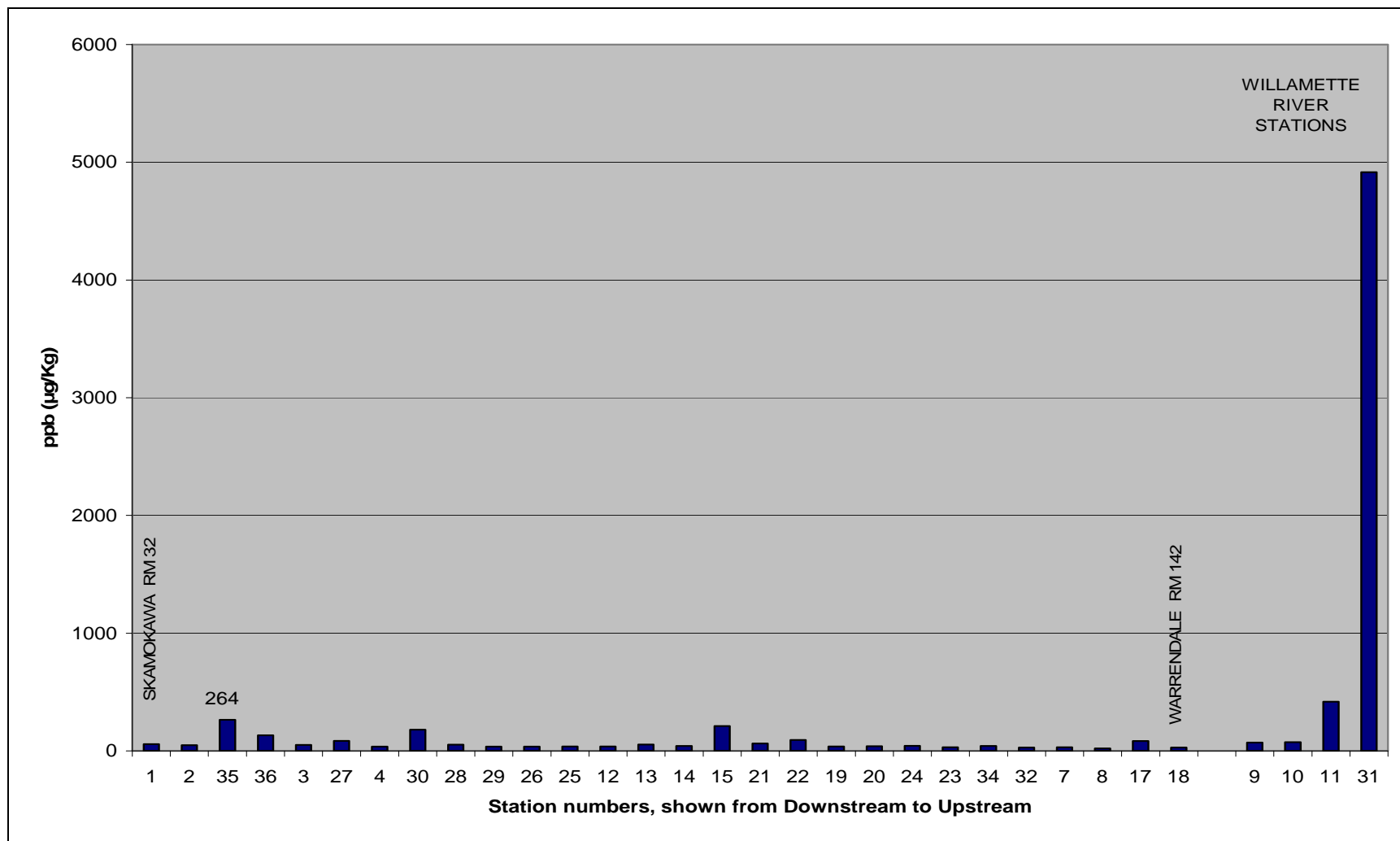


Figure 14. Total PAH in *Corbicula fluminea* by station number in the Columbia and Willamette Rivers.

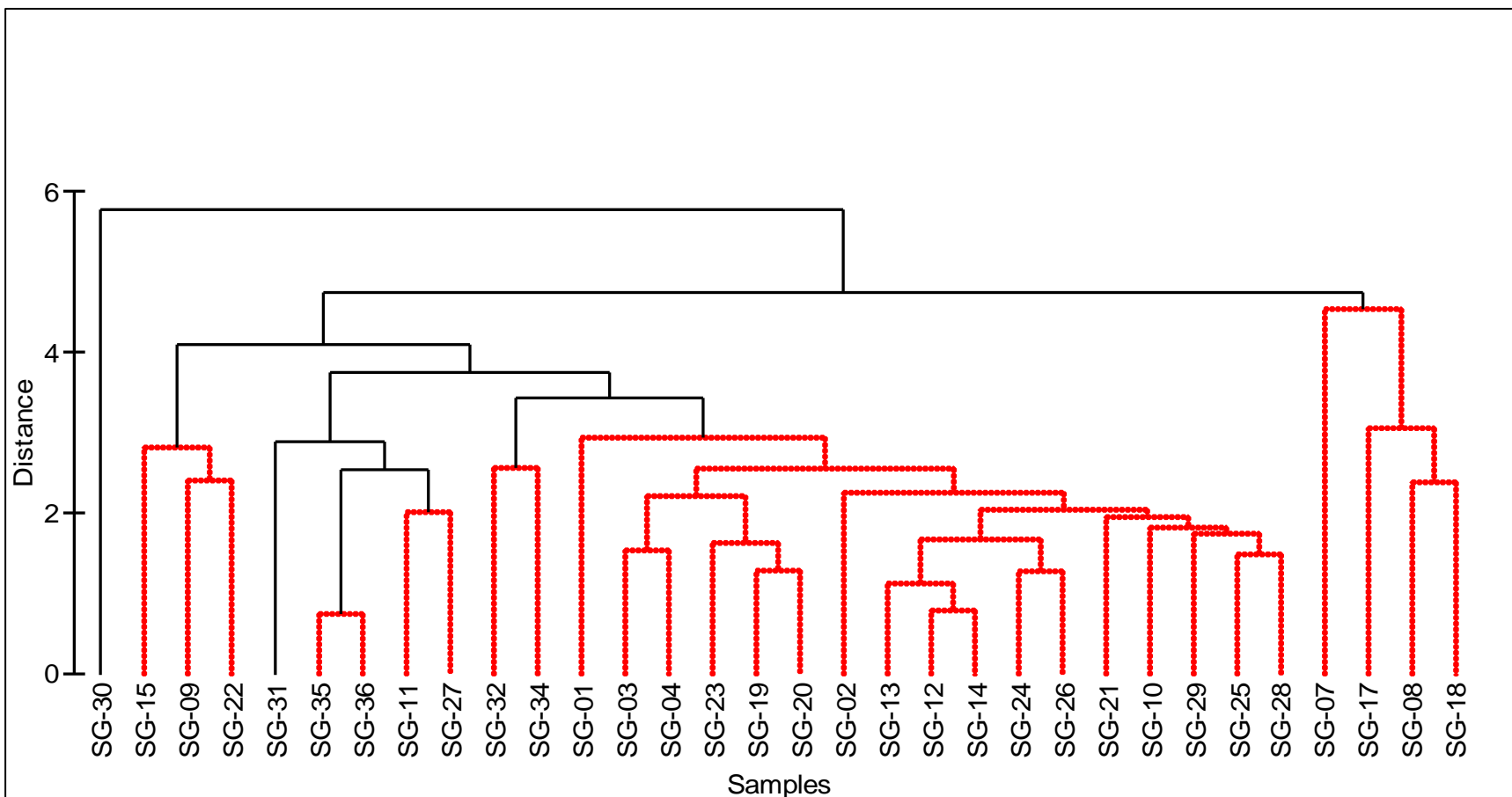


Figure 15. Hierarchical classification of total PAH concentrations in *Corbicula fluminea*.

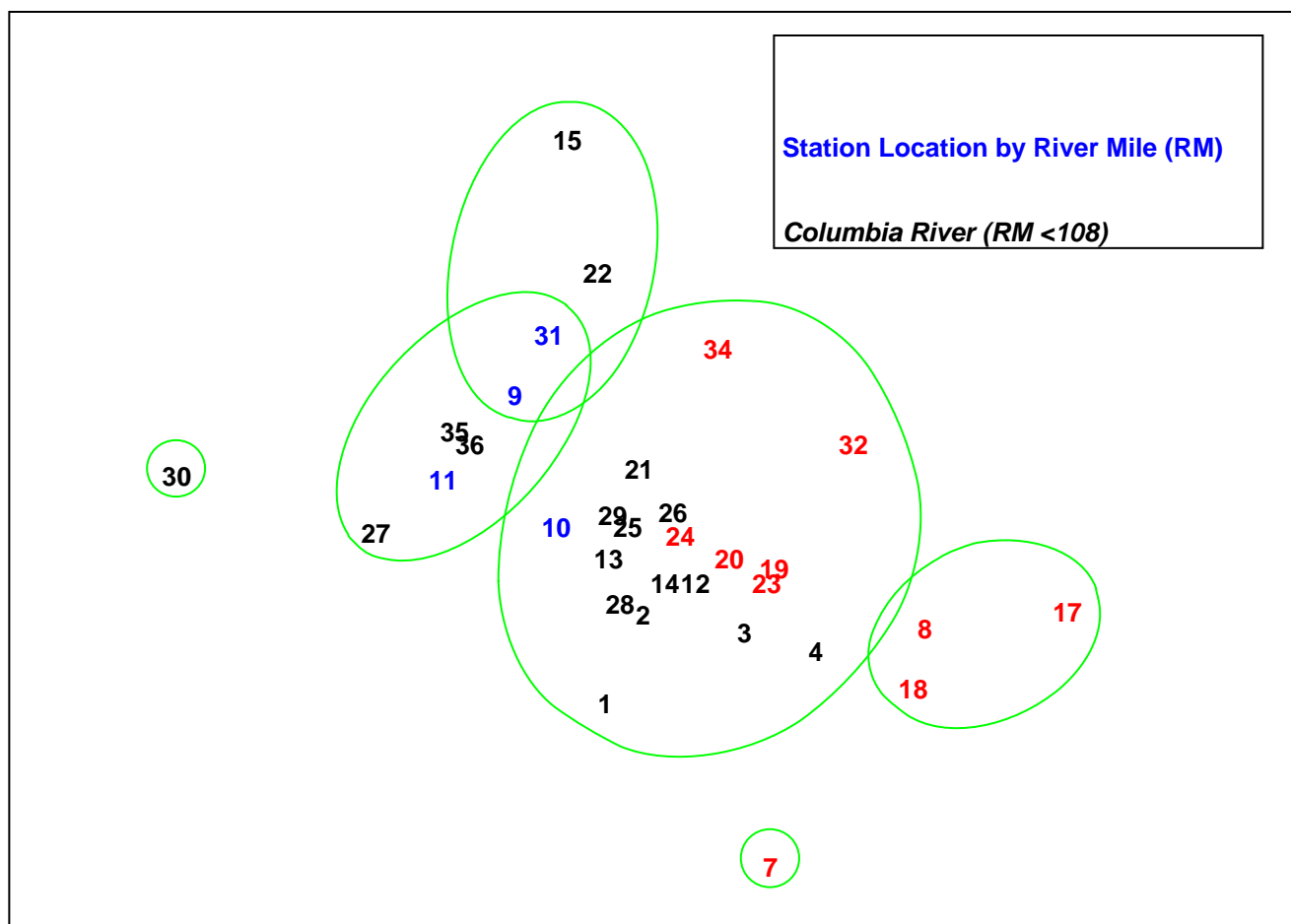


Figure 16. MDS plot for total PAH concentrations in *Corbicula fluminea*. Euclidean distance is represented by green (3.5) lines.

Integration of All Analyte Data for Trends Analysis

Analytical chemistry results for lipid-normalized total PCBs, lipid-normalized total PBDEs, and total PAHs were included in a final analysis to evaluate the potential groupings of sample stations based on chemical profiles for chemicals accumulated in tissues (Figure 17). The analysis separated the groups into four different clusters; characteristics of these groups can be used to infer differences among these clusters. Table 2 displays the nodal analysis with the station groupings separated by double lines, and also the various percentiles of total PAH, PBDE, and PCB concentrations across all stations that were used to color the nodal display. Station 31 stands alone due to the elevated level of PAH. The second group, stations 29 and 30, has the highest levels of PBDE. The third group, stations 13, 12 and 25, has the highest levels of PCBs. The fourth group, stations 8 and 24, has low to very low levels of PAHs and PBDEs, and low to moderate levels of PCBs. The remaining group of 23 stations has variable, but mostly moderate, levels of the three chemical classes.

Table 2. Nodal analysis for lipid-normalized total PCBs and PBDEs, and total PAHs, for all sample stations. The percentiles of the concentrations, displayed at the right, were used to color the cells of the nodal table.

NODAL ANALYSIS FOR ALL DATA (Lipid Normalized)			
Station	PAH ng/g	PBDE ng/g	PCB pg/g
SG-31	4692.50	596.45	2538.77
SG-29	34.81	4044.38	1826.85
SG-30	179.70	6906.92	3371.24
SG-13	52.19	328.53	102441.38
SG-12	35.62	268.13	11925.09
SG-25	37.38	387.92	17777.55
SG-08	20.39	5.42	323.83
SG-24	38.43	42.92	1123.22
SG-26	34.16	1173.64	3032.61
SG-07	30.27	32.16	307.03
SG-11	415.30	486.36	3001.51
SG-22	89.53	173.30	896.26
SG-09	68.66	444.62	2328.97
SG-14	38.61	327.50	1514.41
SG-36	129.31	206.67	530.76

PERCENTILE	PAH	PBDE	PCB
P1	20.39	5.42	307.03
P5	26.1	32.16	313.45
P10	26.89	120.37	413.82
P20	34.56	160.75	530.76
P25	34.76	164.16	535.21
P40	38.1	258.24	852.55
P50	47.63	268.13	1050.72
P60	54.87	327.5	1341.57
P75	84.56	444.62	2328.97

SG-15	208.29	522.19	1341.57		P80	89.53	486.36	2538.77
SG-17	76.06	120.37	313.45		P90	208.29	596.45	3371.24
SG-35	255.56	278.71	852.55		P95	415.3	4044.38	17777.55
SG-04	34.56	267.55	1050.72		P99	4692.5	6906.92	102441.4
SG-18	26.89	188.32	766.54					
SG-21	60.67	410.61	1564.36					
SG-23	26.45	158.56	616.25					
SG-03	48.68	280.80	906.84					
SG-10	72.80	436.75	1526.75					
SG-01	54.87	267.50	535.21					
SG-02	47.63	258.24	769.11					
SG-34	38.10	160.75	452.70					
SG-20	37.72	209.77	548.07					
SG-32	26.10	149.49	413.82					
SG-27	84.56	532.33	1328.06					
SG-19	34.76	164.16	495.80					

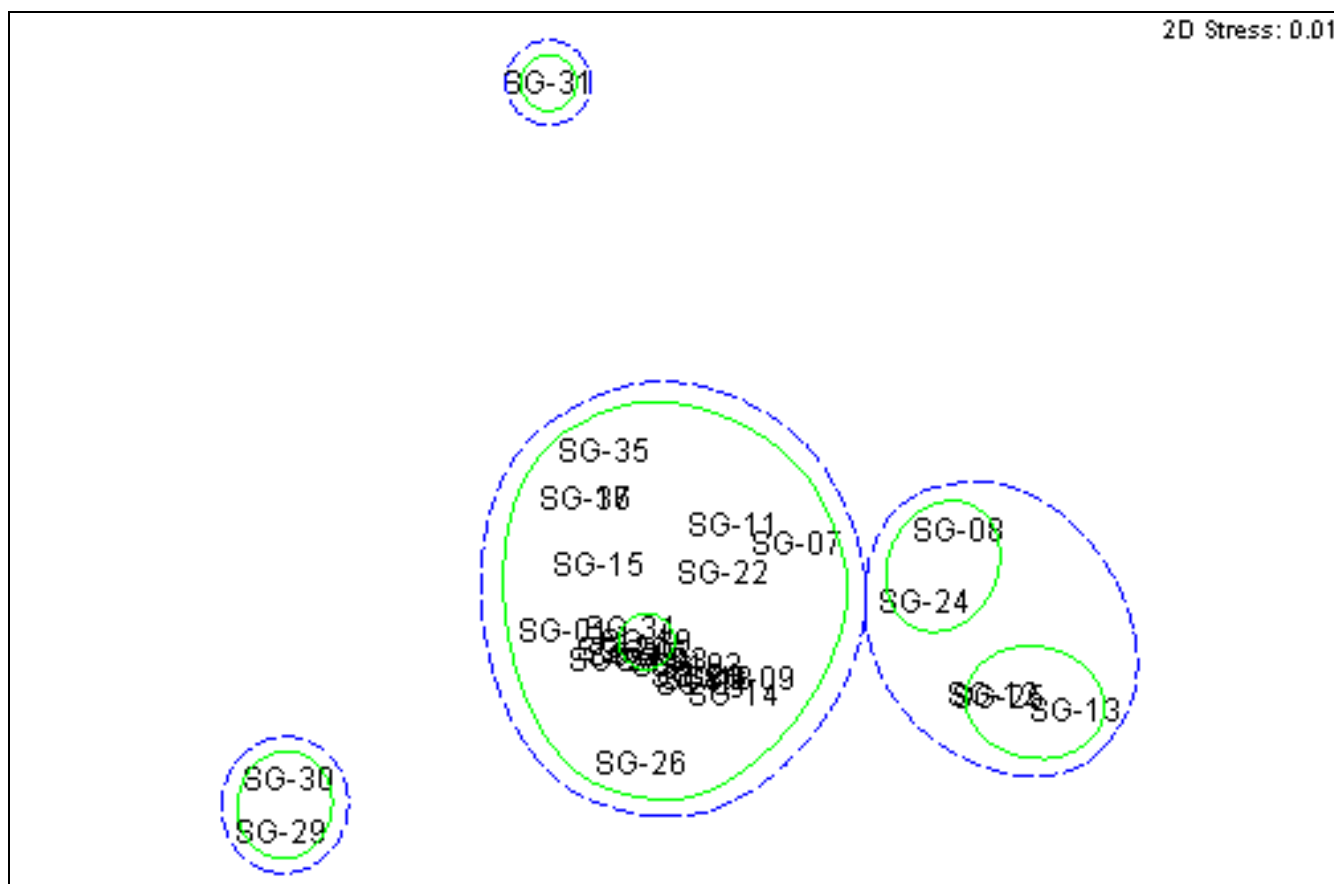


Figure 17. MDS plot for lipid-normalized PCB, lipid-normalized PBDE, and total PAH concentrations in *Corbicula fluminea*. Euclidean distance is represented by green (2.7) and blue (4.1) lines.

4 Discussion and Conclusions

The non-polar organic contaminants including PAHs, PCBs, and PBDEs accumulated and were measured in the tissues of *C. fluminea*. When collected over a large region, such as the Columbia River, the levels of these contaminants in tissues may provide adequate spatial coverage and resolution to be able to discriminate the source and potential exposure routes for these contaminants. Since the spatial scale is on the order of miles and the temporal scale is an average of the life-span of the clams, these values may provide an indication of contaminant bioavailability and bioaccumulation in this system.

More specifically, some of the contaminants were accumulated at much higher levels at some stations than others. For example, PCBs were more readily accumulated at stations 12, 13, and 25 near the confluence of the Willamette River, to levels of 12 to 102 $\mu\text{g/g}$ lipid. Clams in other areas of the river had much lower concentrations, typically 3 $\mu\text{g/g}$ lipid or less. One of the dominant PCBs present in the mixture of congeners was PCB 11, which was found in clams from every station at levels of 14 to 130 ng/g lipid. This PCB, while relatively non-toxic, is rather ubiquitous in urban sediments containing higher chlorinated PCBs and is most likely associated with degradation of PCB-related compounds and has been associated with municipal sewage effluent (Litten et al. 2002).

The flame-retardant compounds, PBDEs, were measured in tissues of *C. fluminea* at all stations, but were the highest at stations 26, 29, and 30, all of which are downstream of the Columbia-Willamette confluence. Generally, the primary PBDE congeners that were present included 47, 100, and 99, in order of their respective concentration at these sites. The same trend for higher levels of PBDE 47 accumulation in tissues was also observed by Svendsen et al. (2007).

The overall distribution of PAH compounds was very heterogeneous; the highest tissue concentration found was on the Willamette River at station 31. This stretch of the Willamette River is known to be highly contaminated with PAHs. As confirmed by the nodal analysis in Table 2, the sites

with the highest levels of tissue-associated contaminants were stations in or near the Portland-Vancouver metropolitan area.

Tissue concentrations in *C. fluminea* were compared to data for other organisms and surrogates that have been collected in the Columbia River, including gut contents of field-collected downstream migrating juvenile salmonids (LCREP 2007), and semi-permeable membrane device (SPMD) extracts (Johnson 2005). All three independent studies were conducted in 2005. Figures 18-20 show a compilation of data for clam tissue, fish gut content, and SPMDs collected over a similar distance of the Columbia and Willamette Rivers for PAHs, PCBs, and PBDEs. SPMDs were deployed at five sites along the mainstem Columbia in April and August of 2005. Gut contents of downstream migrating juvenile Chinook salmon were collected at six sites from April to August 2005. Clams, SPMDs, and fish gut contents were analyzed for PCBs (Figure 18) and PAHs (Figure 19). In addition, clams and fish gut content were analyzed for PBDEs (Figure 20).

To evaluate the similarity of measures using these three different approaches, the response magnitude was plotted against river mile on the Columbia and Willamette Rivers. While the concentrations cannot be compared directly (i.e., due to lipid normalization and matrix), the relative response can be used to indicate increased relative exposure in specific regions of the river. The other measures had trends of elevated contaminants near or around or downstream of the Portland-Vancouver metropolitan area similar to the *C. fluminea* data in this study. Fish gut content concentrations appear to have closer specific spatial trends to the clam data than the SPMDs. Higher levels in gut content of PAHs and PBDEs appear to have a downstream lag when compared to the highest levels found in the clam data. This may be due primarily to the migratory behavior of the salmon smolts that were sampled. The pattern of concentrations in the SPMDs located in the mainstem of the Columbia River do not show these spatial trends but appear uniform over the sampling area in comparison to the clam and gut content data.

Trends in PCB clam and salmon gut content are similar though the magnitude of contamination can differ near known sources. Near River Mile 101-103 a significant spike in PCB was observed for *C. fluminea*; this is also the location of highest PCB contamination in salmon stomach content collected at RM 101. Where clam and salmon stomach content

stations are synoptic, the concentrations in the clams are consistently higher. However, except for the high spike in the clam data at RM 103, they are generally in the same order of magnitude. Clams, if used as a surrogate for migrating juvenile salmon, would overestimate contaminant exposure.

The SPMD data for PCBs as for PAHs and PBDEs show more variability between water bodies than between the various locations of the mainstem Columbia. This is likely due to the association of the SPMDs with contaminants found in the water column as the SPMDs were suspended in the water column and are not in direct contact with sediment. Small localized contaminant sources do not appear to be captured by SPMDs in areas of high river flows.

In the current study, these data provide additional information regarding the performance and capacity for field-collected clams to provide information regarding the uptake and spatial distribution of sediment-associated contaminants. In specific evaluations, such as a site assessment for remediation or dredging, in situ clam tissue data could be used as an additional line of evidence for the assessment of potential bioaccumulation, as surrogates for species of concern, as well as empirical data to strengthen food web models.

Field-collected data, such as tissue residues in *C. fluminea*, may provide significant value as empirical data for food web models. These models rely on site-specific data such as sediment concentrations, invertebrate concentrations from laboratory bioassays, or field-collected concentrations in fish. These measures have their limitations including possible inability to predict bioaccumulation from sediments, potential that steady-state concentrations will not be reached during bioassays, lack of invertebrate models relevant to the exposure system, or migratory nature of the animals sampled. In the context of an ecological risk assessment, *C. fluminea* may provide a direct measure and increased resolution of temporal and spatial variability over a large system as well as serving as a representative or actual prey species within the conceptual site model. On a large scale, such as the Columbia River, field-collected clams can be used as a tool for identifying regions of the river that may have distinct contaminant patterns or elevated levels of contaminants. Analytical chemistry results from field-collected clams may be used as part of a

bioaccumulation assessment as outlined in the simplified conceptual model shown in Figure 21. The conceptual model for *C. fluminea* has three main components; exposure, accumulation, and trophic transfer.

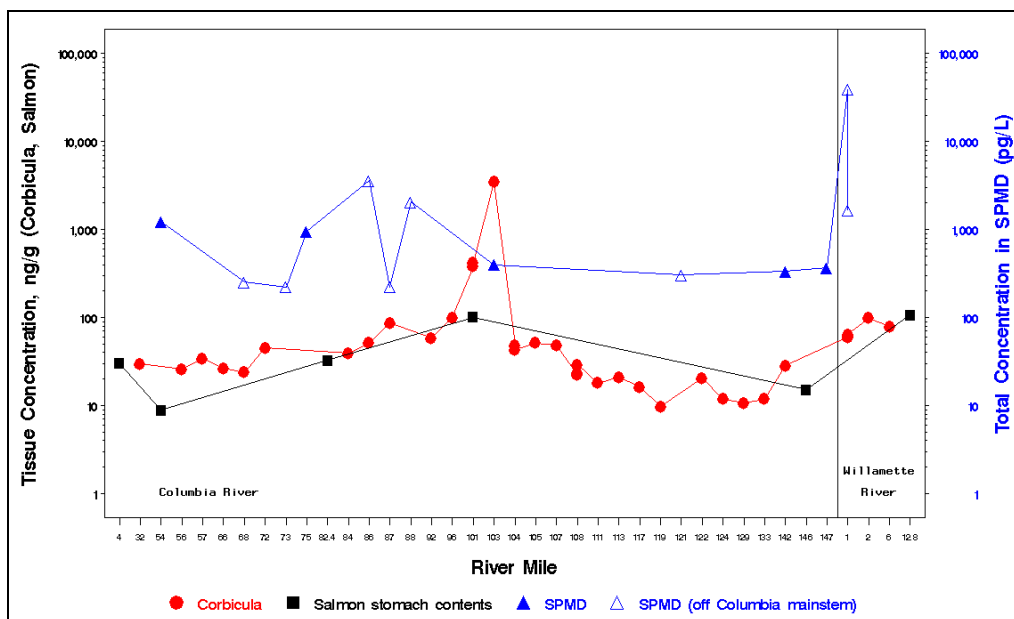


Figure 18. Sum of PCB compounds in *Corbicula fluminea*, stomach contents in field-collected salmon, and concentrations observed in SPMDs.

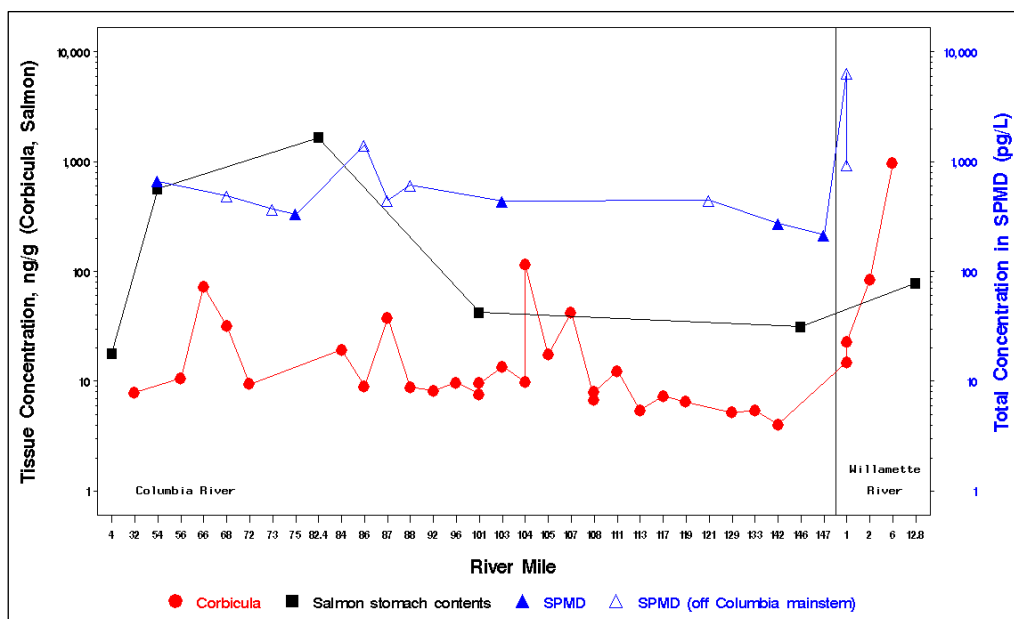


Figure 19. Sum of PAH compounds in *Corbicula fluminea*, stomach contents in field-collected salmon, and concentrations observed in SPMDs. Sum of PAH was determined by totaling concentrations of benzo[a]pyrene, benz[a]anthracene, benzo[b]fluoranthene, benzo[k]fluoranthene, benzo[ghi]perylene, chrysene, dibenz[a,h]anthracene and indeno[123-cd]pyrene.

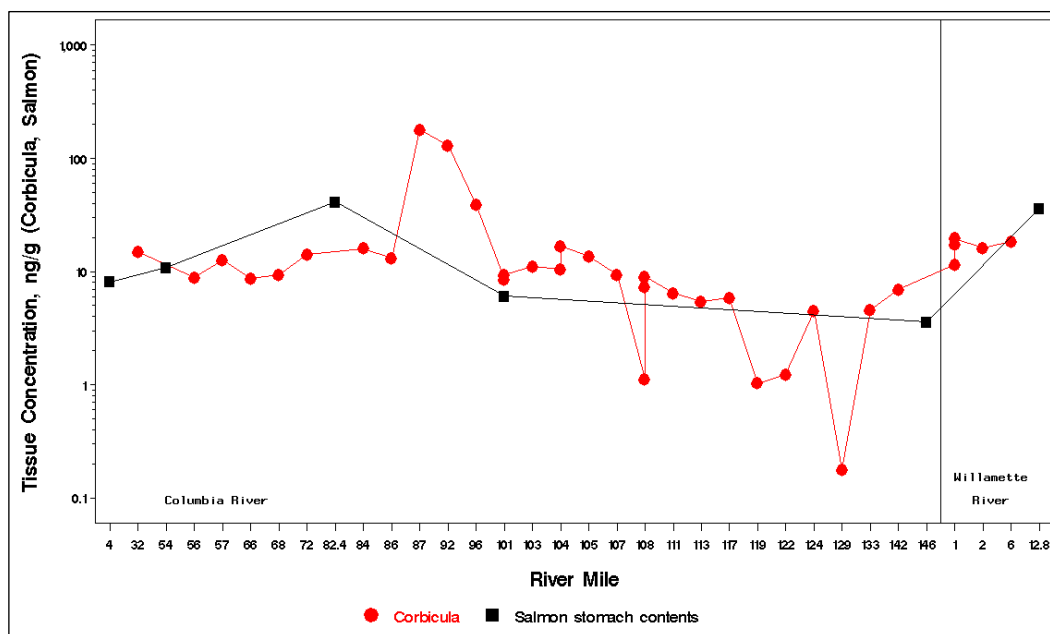


Figure 20. Sum of PBDE compounds in *Corbicula fluminea* and stomach contents in field-collected salmon.

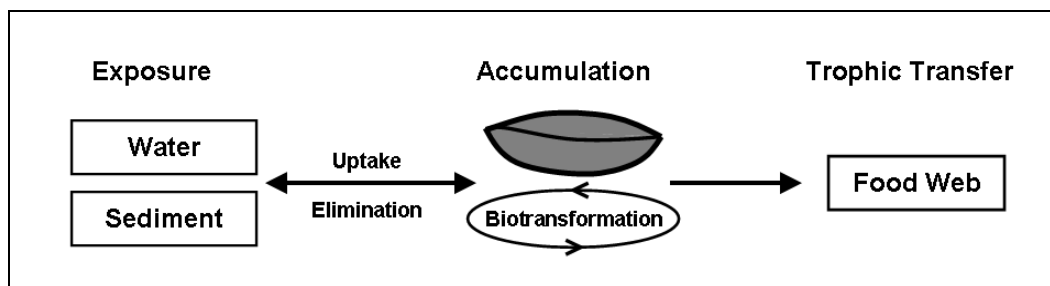


Figure 21. Simplified conceptual model for exposure, uptake, and trophic transfer of contaminants using field-collected *C. fluminea* tissue concentrations.

The accumulation of contaminants by *C. fluminea* is expected to occur primarily through deposit feeding or pedal feeding and filter feeding; although pedal feeding is the dominant mechanism of feeding (Vaughn and Hakenkamp 2001). Water quality, nutrients, physical disturbance, and grain size may modify the proportion of food and subsequently contaminants that may be taken up from sediment or water through pedal feeding or filter feeding, respectively (Hakenkamp et al. 2001). The processes responsible for these behavior modifications and differences in uptake mechanisms must be resolved or better understood prior to application within bioaccumulation assessment and food web modeling. For example, how do *C. fluminea* concentrations from field-collected organisms compare to those exposed to a contaminant in a standardized

28-day bioassay? Data from the Lower Willamette Group report (USEPA 2007a) suggest field and laboratory tissue concentrations will be much different. These differences may be due to the inability to reach steady state in bioassay conditions, variability of exposure concentrations, different behavior of animals in bioassay versus field (e.g., clamming up in bioassay), or filter feeding behavior of clams (versus pedal feeding).

Accumulation of contaminants occurs when the rate of contaminant uptake exceeds contaminant elimination. While uptake is controlled primarily by contaminant bioavailability and feeding, elimination is controlled by biotransformation and excretion. Metabolism of halogenated organic compounds such as organochlorine pesticides, PCBs, and PBDEs is expected to be very limited. However, some evidence suggests that bivalves have a high potential for metabolism of PAHs (Livingstone et al. 1989). In a study by Narbonne et al. (1999) very limited metabolism and elimination were observed in *C. fluminea* for four PAH compounds (anthracene, phenanthrene, benzo[a]pyrene and pyrene). The current study appears to further demonstrate the potential for *C. fluminea* to accumulate PAH compounds.

The use of *C. fluminea* as a measure of contaminant accumulation and trophic transfer within the aquatic food web can support comprehensive food web modeling. Clams and other large invertebrates are prey for certain species of fish, mammals, and birds. In some systems, such as the Columbia River, *C. fluminea* are directly consumed by demersal fish including sturgeon (Mason and Clugston 1993). However, *C. fluminea* are unlikely to be consumed by species such as juvenile Chinook salmon that eat primarily larval fish and amphipods (Schabetsberger et al. 2003). In these cases, *C. fluminea* may be used as a surrogate or indicator species of the level of contaminant accumulation that may occur in other benthic prey.

The statistical tools used in this study, multidimensional scaling, nodal analysis, and hierarchical classification, are methods that have historically been used to evaluate differences among sampling sites based on the presence, diversity, and distribution of benthic invertebrate species. Application to a data set of contaminant concentrations and sampling locations provides an opportunity to identify sites with similar contaminant profiles, characteristics, and levels. These multivariate techniques can be used

alone to identify “hot spot” contamination or in combination with advanced geographical information systems (GIS) to gain a better understanding of the spatial distribution of contaminants within a large system such as the Columbia River.

The use of *C. fluminea* for monitoring, site assessment, or sediment evaluations appears to be promising, and additional studies should be conducted to relate this measure to other existing measures of exposure and contaminant uptake. Other studies that have been conducted on sediments and contaminants from the Columbia River may provide additional information on the utility of *C. fluminea* as a measure of bioaccumulation. These studies include the EPA’s Lower Willamette Group report (USEPA 2007a) that describes contaminant accumulation in *L. variegatus* and *C. fluminea* in laboratory bioassays, as well as field-collected clams, crayfish, and sculpin. Two studies on the Willamette River conducted paired laboratory bioaccumulation using *L. variegatus* and *C. fluminea* (USACE 2002, 2004). These studies, while focused on a smaller region of the watershed, also provide the basis for assessing the predicting capability of *C. fluminea* to other species. These studies should be further examined using the data in the current study to gain a better understanding of the processes that are important for the bioaccumulation of organic contaminants in the Columbia River system.

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Appendix A: Chemistry Database

Water Body	Sample Identifier	Station	River Mile	Latitude	Longitude	Sample Location	State
Columbia River	CR32-SG-01	SG-01	32	46.26827778	-123.4598333	Skamokawa_Vista_Park	WA
Columbia River	CR56-SG-02	SG-02	56	46.18980556	-123.12575	Stella	WA
Columbia River	CR57-SG-16	SG-16	57	46.17688889	-123.0986389	Willow_Grove	WA
Columbia River	CR66-SG-35	SG-35	66	46.11119444	-122.9643611	Longview_WA_West_of_CR_Bridge	WA
Columbia River	CR68-SG-36	SG-36	68	46.09627778	-122.9286389	Old_Mouth_of_the_Cowlitz	WA
Columbia River	CR75-SG-03	SG-03	75	46.05283	-122.8736	Kalama	WA
Columbia River	CR84-SG-27	SG-27	84	45.87319444	-122.7951667	St_Helens_Sand_Island	OR
Columbia River	CR86-SG-04	SG-04	86	45.87031	-122.77869	Woodland	WA
Multnomah Channel	CR87-SG-30	SG-30	87	45.85275	-122.7948333	Mouth_of_Multnomah_Channel	OR
Lake River	CR88-SG-28	SG-28	88	45.84072222	-122.7778333	Mouth_of_Lake_River	WA
Columbia River	CR92-SG-29	SG-29	92	45.79361111	-122.7766389	Head_of_Bachelor_Slough	WA
Columbia River	CR96-SG-26	SG-26	96	45.73392	-122.75591	End_of_NW_Lower_River_Road	WA
Columbia River	CR101-SG-12	SG-12	101	45.66008333	-122.7567778	WA_side_opposite_Willamette_River_Mouth	WA
Columbia River	CR101-SG-25	SG-25	101	45.6659	-122.7606	Vancouver_Flushing_Channel	WA
Columbia River	CR103-SG-13	SG-13	103	45.6465	-122.7379722	Vanalco	WA
Columbia River	CR104-SG-14	SG-14	104	45.64208333	-122.7198333	No_name_Washington_Shore	WA
Columbia River	CR104-SG-15	SG-15	104	45.63541667	-122.7227222	Hayden_Island	OR
Columbia River	CR105-SG-21	SG-21	105	45.62386111	-122.6965278	Hayden_Island	OR
Columbia River	CR107-SG-22	SG-22	107	45.61488889	-122.6769722	Under_South_end_of_I-5_Bridge	OR
Columbia River	CR108-SG-19	SG-19	108	45.60522222	-122.6494722	Tomhawk_Island_Columbia_River	OR
Columbia River	CR108-SG-20	SG-20	108	45.60394444	-122.65225	Tomhawk_Island_Oregon_Slough	OR
Columbia River	CR108-SG-24	SG-24	108	45.611747	-122.634975	Vancouver_Historic_Site Boatramp	WA
Columbia River	CR113-SG-23	SG-23	110	45.61181	-122.60899	Wintler_Park_Vancouver	WA
Columbia River	CR111.5-SG-34	SG-34	111.5	45.59861111	-122.5785278	Lemon_Island_DS	OR
Columbia River	CR112.5-SG-33	SG-33	112.5	45.58855556	-122.5497222	Government_Island_DS_of_I-205	OR
Columbia River	CR117-SG-32	SG-32	117	45.57875	-122.4712222	Sand_Island	OR
Columbia River	CR119-SG-07	SG-07	119	45.57197222	-122.4387778	Lady_Island	WA
Columbia River	CR122-SG-06	SG-06	122	45.57222222	-122.3785	Shallow_Water_across_from_Camas	OR
Columbia River	CR124-SG-05	SG-05	124	45.56358333	-122.3368611	Downstream_Reed_Is	WA
Columbia River	CR129-SG-08	SG-08	129	45.54769444	-122.23975	Rooster_Rock	OR
Columbia River	CR133-SG-17	SG-17	133	45.55994444	-122.1784722	Bridal_Veil	OR
Columbia River	CR142-SG-18	SG-18	142	45.61502778	-122.0028056	Warrendale	OR
Willamette River	WR1E-SG-09	WR-09	1E	45.64472222	-122.7685278	Willamette_River_DS_of_Columbia_Slough	OR
Willamette River	WR1W-SG-10	WR-10	1W	45.64825	-122.7720278	Willamette_River_across_from_Columbia_Slough	OR
Willamette River	WR1W-SG-10A	WR-10A	1W	45.64825	-122.7720278	Duplicate	OR
Willamette River	WR2-SG-11	WR-11	2W	45.62466667	-122.79475	Sauvie_Island	OR
Willamette River	WR6.5-SG-31	WR-31	6W	45.58125	-122.7639722	US_Moorings	OR

Appendix B: Data Validation

In August and September 2005, a total of 36 nearshore locations were sampled for *Corbicula fluminea*. Tissue samples were analyzed for the following bio-accumulative constituents: semi-volatile compounds (including PAHs), chlorinated pesticides, PCB aroclor mixtures, and all 209 congeners, polybrominated diphenyl ethers (PBDE; fire retardants), organotins, and metals (mercury (Hg), lead (Pb), zinc (Zn), cadmium (Cd)). The analytical methods used by the laboratories are shown in Table B1.

Table B1. Analytical methods

Analytes	Analytical Method
PCB Aroclor mixtures	EPA SW846 8082
PCB Congeners (209)	EPA SW846 1668A
PBDE	EPA SW846 1614
Semi-volatiles	EPA SW846 8270C
PAHs	EPA SW846 8270C
Mercury	EPA SW846 7471
Metals (Pb, Zn, Cd)	EPA SW846 6020
Organotin	NMFS KRONE Method
Lipids	EPA SW846 8290
Pesticides	EPA SW846 8081

All samples were sent to STL Sacramento for analyses. STL Sacramento performed all analyses except organotin, semi-volatiles, and PAHs. The organotin analyses were subcontracted to STL Burlington, Indiana. The semi-volatiles and PAH analyses were subcontracted to CAS Kelso in Longview, Washington.

Table B2. Listing of all sample identifications, dates of collection, and analytical methods performed.

USACE Sample Identification	Sample Collection Date	PCBs Aroclor Mixtures	PCBs Congeners	PBDE	Semi-volatiles	PAHs	Mercury	Metals (Cd, Pb, Zn)	Organotin	Lipids	Pesticides
080405CR32-SG-01	8/4/05 10:00 AM	•	•	•	•	•	•	•	•	•	•
080405CR56-SG-02	8/4/05 11:52 AM	•	•	•	•	•	•	•	•	•	•
080405CR75-SG-03	8/4/05 1:25 PM	•	•	•	•	•	•	•	•	•	•
080505CR86-SG-04	8/5/05 2:00 PM	•	•	•	•	•	•	•	•	•	•
083105CR124-SG-05	8/31/05 10:35 PM	•	•	•	•		•	•	•	•	•
083105CR122-SG-06	8/31/05 11:58 PM	•	•	•	•		•	•	•	•	•
083105CR119-SG-07	8/31/05 12:35 PM	•	•	•	•	•	•	•	•	•	•
083105CR129-SG-08	8/31/05 1:45 PM	•	•	•	•	•	•	•	•	•	•
090805WR1E-SG-09	9/8/05 8:45 AM	•	•	•	•	•	•	•	•	•	•
090805WR1W-SG-10	9/8/05 9:45 AM	•	•	•	•	•	•	•	•	•	•
090805WR1W-SG-10A	9/8/05 9:45 AM	•	•	•			•	•	•	•	•
090805-WR2-SG-11	9/8/05 11:01 AM	•	•	•	•	•	•	•	•	•	•
090805-CR101-SG-12	9/8/05 11:32 AM	•	•	•	•	•	•	•	•	•	•
090805-CR103-SG-13	9/8/05 12:25 AM	•	•	•	•	•	•	•	•	•	•
090805-CR104-SG-14	9/8/05 1:21 PM	•	•	•	•	•	•	•	•	•	•
090805-CR104-SG-15	9/8/05 1:51 PM	•	•	•	•	•	•	•	•	•	•
091505CR57SG-16	9/15/05 9:45 AM	•	•	•			•	•	•	•	•
092105CR133-SG-17	9/21/05 8:00 AM	•	•	•	•	•	•	•	•	•	•
092105CR142-SG-18	9/21/05 12:00 PM	•	•	•	•	•	•	•	•	•	•
092205CR108-SG-19	9/22/05 8:25 AM	•	•	•	•	•	•	•	•	•	•

Table B2											
USACE Sample Identification	Sample Collection Date	PCBs Aroclor Mixtures	PCBs Congeners	PBDE	Semi-volatiles	PAHs	Mercury	Metals (Cd, Pb, Zn)	Organotin	Lipids	Pesticides
092205CR108-SG-20	9/22/05 10:05 AM	•	•	•	•	•	•	•	•	•	•
092205CR108-SG-20A	9/22/05 10:05 AM	•					•	•	•	•	•
092205CR105-SG-21	9/22/05 11:00 AM	•	•	•	•	•	•	•	•	•	•
092205CR107-SG-22	9/22/05 12:20 PM	•	•	•	•	•	•	•	•	•	•
092605CR113SG-23	9/26/05 10:15 AM	•	•	•	•	•	•	•	•	•	•
092605CR108SG-24	9/26/05 11:20 AM	•	•	•	•	•	•	•	•	•	•
092605CR101SG-25	9/26/05 1:15 PM	•	•	•	•	•	•	•	•	•	•
092605CR96SG-26	9/26/05 2:25 PM	•	•	•	•	•	•	•	•	•	•
092705CR84SG-27	9/27/05 10:25 AM	•	•	•	•	•	•	•	•	•	•
092705CR88SG-28	9/27/05 11:20 AM				•	•					•
092705CR92SG-29	9/27/05 12:35 PM	•	•	•	•	•	•	•	•	•	•
092705CR87SG-30	9/27/05 1:30 PM	•	•	•	•	•	•	•	•	•	•
092805 WR6.5-SG-31	9/28/05 9:40 AM	•	•	•	•	•	•	•	•	•	•
092805 CR 117-SG-32	9/28/05 12:40 PM	•	•	•	•	•	•	•	•	•	•
092805 CR 111-SG-34	9/28/05 1:30 PM	•	•	•	•	•	•	•	•	•	•
092905 CR 66-SG-35	9/29/05 10:00 AM	•	•	•	•	•	•	•	•	•	•
092905 CR 68-SG-36	9/29/05 10:31 AM	•	•	•	•	•	•	•	•	•	•

Quality Assurance Review

Upon receipt of the laboratory reports, the analytical results were reviewed based on the following quality control (QC) measures, as appropriate:

- Sample holding times
- Blank analysis results
- Surrogate recoveries
- Analytical sequence
- Sample condition upon laboratory receipt
- Initial and continuing calibration verification standards
- Laboratory control sample/laboratory control sample duplicate (LCS/LCSD) recoveries and precision
- Quantitation results
- Matrix spike/matrix spike duplicate (MS/MSD) recoveries and precision
- Dual column chromatographic precision

Validation of the analytical results is based on an evaluation of the data deliverables provided by the laboratories. Review of the data has been performed based on the “National Functional Guidelines for Organic Data Review” (USEPA 2007b) and the “National Functional Guidelines for Inorganic Data Review” (USEPA 2007c). It should be noted that the National Functional Guidelines specifically address analyses performed in accordance with the Contract Laboratory Program (CLP) analytical methods and are not completely applicable to the type of analyses and analytical protocols performed for the SW-846 and USEPA Methods utilized by the laboratory for these samples. Professional judgment was used to determine the usability of the analytical results and compliance relative to the SW-846 and USEPA Methods utilized by the laboratory. Validation flags were not added to the analytical results.

PCB Aroclor Mixtures

The laboratory chose to quantify detected Aroclor mixtures as the most prevalent Aroclor rather than attempting to quantify concentrations of multiple Aroclors in a single sample. The identity of the Aroclor in these determinations may be misleading. Quality control analyses indicate that analytical conditions were within control for these samples. Some Aroclors were quantified as estimates by the laboratory, since they were detected between laboratory detection limits and reporting limits. A few samples

were analyzed at a dilution due to the high concentration of analytes. Reporting limits were adjusted accordingly. Aroclor data are considered usable for these samples with the caveat that not all Aroclors were quantified separately in each sample.

PCB Congeners

Due to matrix interference, as indicated by out-of-control matrix spike and matrix spike duplicate results, congener analyses may have a high bias. Some surrogate recoveries exceeded control limits. Some congener results were flagged by the analytical laboratory indicating that the measured concentration exceeded the instrument calibration range while not reaching detector saturation. These values should be considered estimates. Based on laboratory experience, which is detailed in the laboratory report case narrative, re-analyzing samples at a dilution (which would have brought the result into calibration range) would not provide substantially different results. PCB 180 was detected in the method blank associated with laboratory batch 5266259. Samples analyzed in this batch include those listed below:

090805WR1E-SG-09	090805-CR101-SG-12
090805WR1W-SG-10	090805-CR103-SG-13
090805WR1W-SG-10A	090805-CR104-SG-14
090805WR2-SG-11	090805-CR104-SG-15

PCB congener data for all samples are considered usable.

PBDE

Based on some spiking compound recoveries out of control high in the laboratory control, matrix spike and matrix spike duplicate analyses, a potential high bias attributed to matrix interference and laboratory performance is indicated for the following samples:

090805WR1E-SG-09	091505CR57SG-16
090805WR1W-SG-10	092605CR113SG-23
090805WR1W-SG-10A	092605CR108SG-24
090805-WR2-SG-11	092605CR101SG-25
090805-CR101-SG-12	092605CR96SG-26
090805-CR103-SG-13	092705CR84SG-27
090805-CR104-SG-14	092705CR92SG-29
090805-CR104-SG-15	092705CR87SG-30

PBDE data are considered usable for all samples.

Semi-Volatiles

Based on LCS performance, benzoic acid should be considered a low estimate in most samples. Phenol and benzoic acid were detected at estimated levels below reporting limits in method blanks. Results for phenol and benzoic acid were flagged by the laboratory, indicating method blank contamination. A few samples exhibited poor internal standard recoveries. There is a potential high bias for pentachlorophenol as indicated by matrix spike performance for the following samples:

092105CR133-SG-17	092705CR84SG-27
092105CR142-SG-18	092705CR88SG-28
092205CR108-SG-19	092705CR92SG-29
092205CR108-SG-20	092705CR87SG-30
092205CR105-SG-21	092805 WR6.5-SG-31
092205CR107-SG-22	092805 CR 117-SG-32
092605CR113SG-23	092805 CR 111-SG-34
092605CR108SG-24	092905 CR 66-SG-35
092605CR101SG-25	092905 CR 68-SG-36
092605CR96SG-26	

All semi-volatile data are considered usable with the possibility of a potential high bias for pentachlorophenol in some samples and potential low bias for benzoic acid in most samples.

PAHs

Results for naphthalene, dibenzofuran, phenanthrene, 2-methylnaphthalene, acenaphthylene, and fluorine could be elevated due to laboratory contamination as indicated by the method blank results. The laboratory noted method blank contamination with the appropriate flag for these samples. All data are considered usable.

Mercury

All mercury data for the *Corbicula fluminea* samples should be considered low estimates with possible false negatives. This assessment is based on low recoveries for the matrix spike and matrix spike duplicate analyses in multiple batches of samples. All data are considered usable with the caveat that the mercury results should be considered low estimates.

Metals (Cd, Pb, Zn)

Lead was detected at estimated levels between the reporting limit and detection limit in the method blank for one batch of samples and zinc in another. Potentially this could indicate that lead and zinc concentrations in the samples could have a high bias. Ultimately it was determined that the lead and zinc method blank concentrations were not significant. All data are considered usable.

Organotin

Tributyltin was detected in some samples and flagged by the laboratory, indicating greater than a 25-percent difference between dual column quantitation. The laboratory noted that the lower of the two values was reported. These results should be viewed with caution. Due to insufficient sample, most analytical batches of samples did not include matrix spike or matrix spike duplicate analyses. The matrix spike and matrix spike accuracy and precision were within laboratory control limits for the one group of samples that included them. Accuracy for all LCS determinations with all sample groups was within laboratory control limits. Organotin data are considered usable.

Lipids

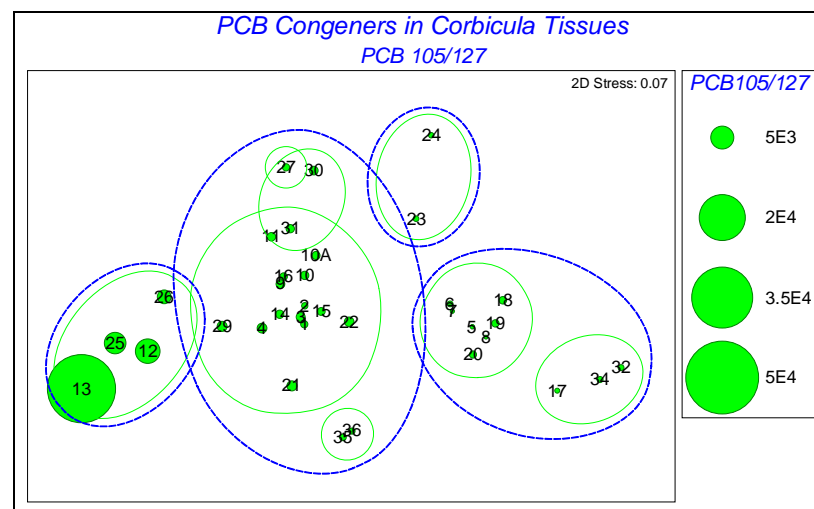
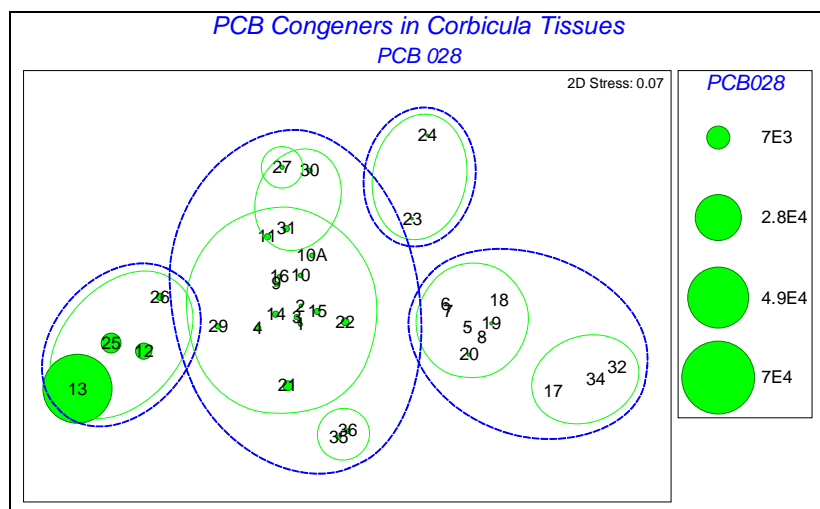
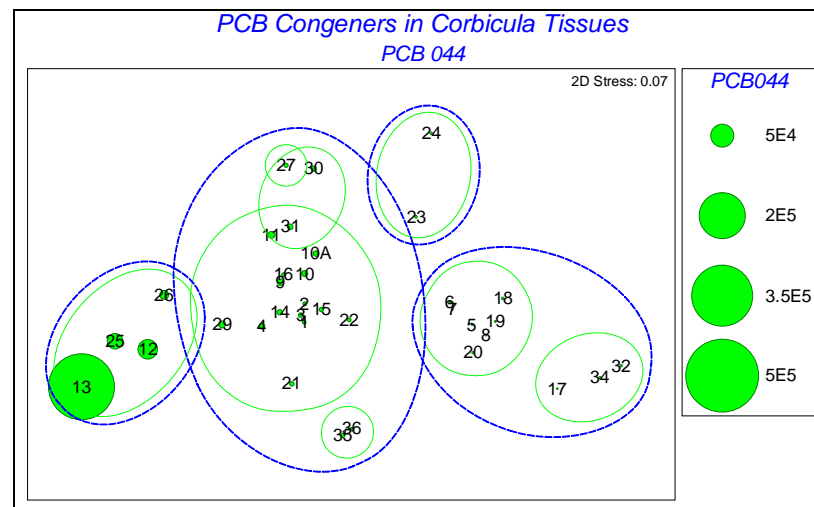
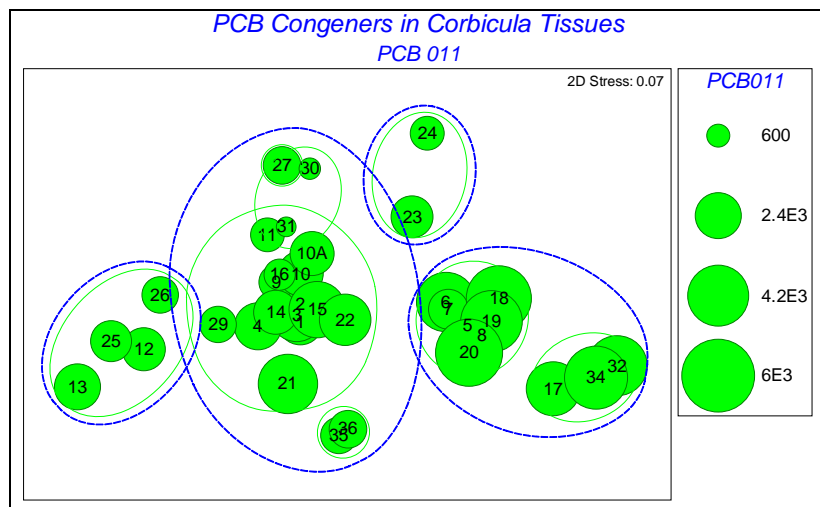
No anomalies were noted with this data.

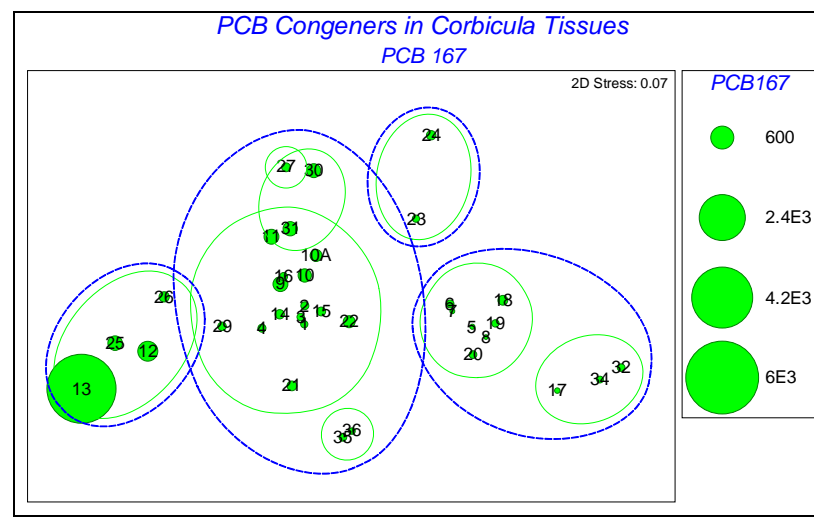
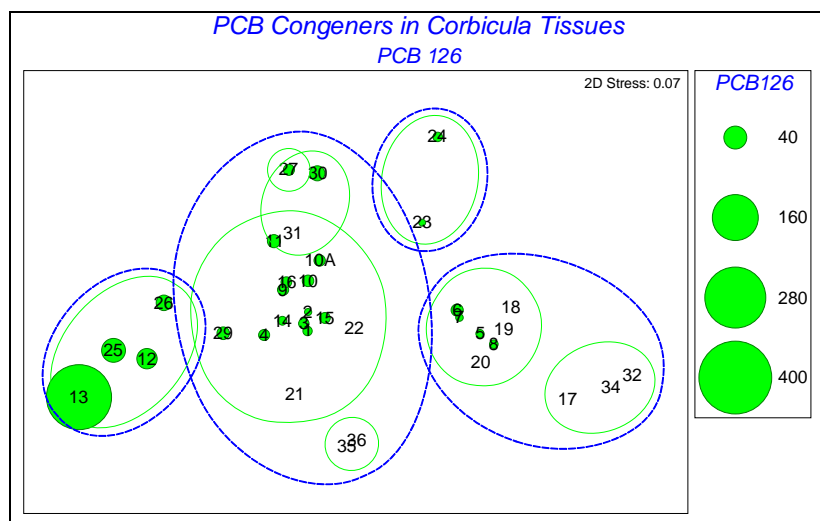
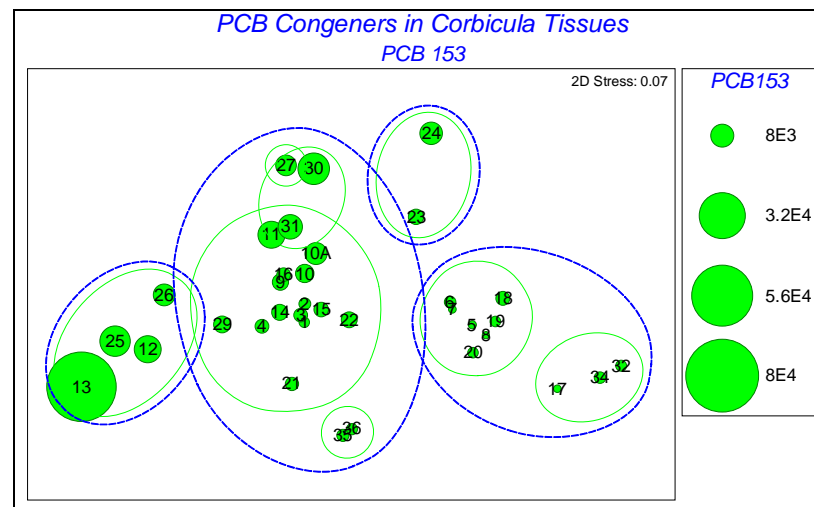
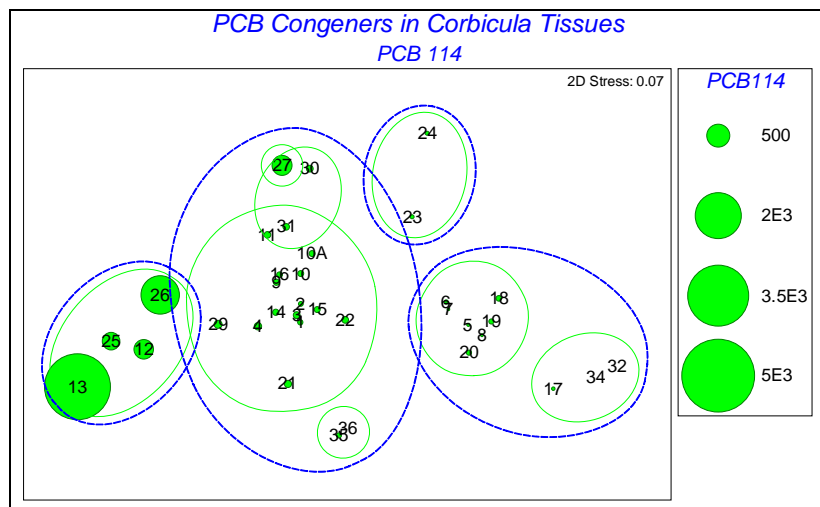
Pesticides

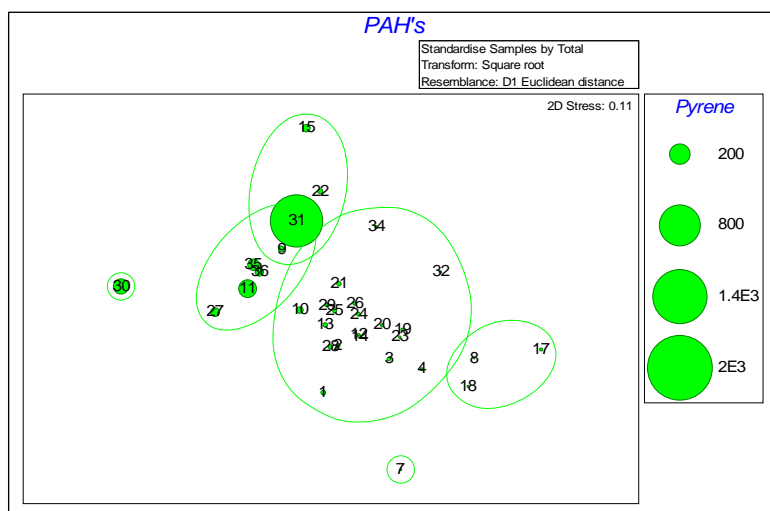
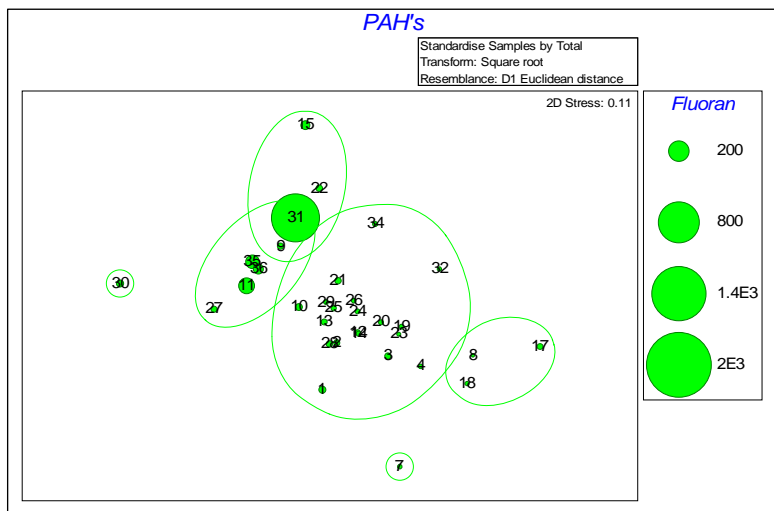
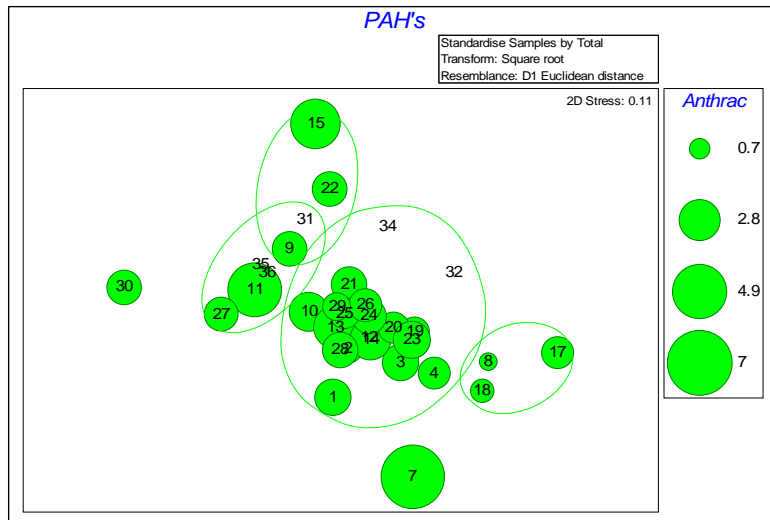
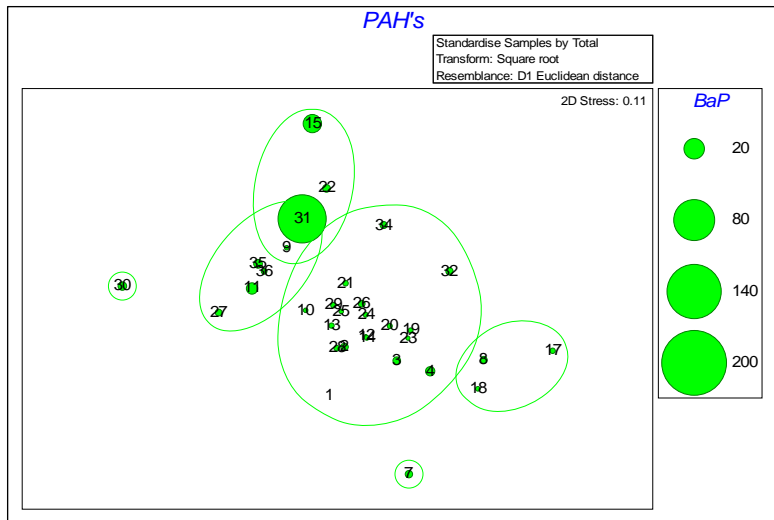
All pesticide 8081 data should be considered tentatively identified and should be considered estimates with possible false negatives. Method 8081 relies on dual-column injection and second-column confirmation for identification and quantitation of analytes. As indicated by deteriorating continuing calibration verification (CCV) analyses and lack of second column confirmation, matrix effects caused difficulty with this analysis. Since the analytes in the samples could not be confirmed on the second column and the CCV analyses on the primary and secondary columns progressively failed over the course of the analytical run, the pesticide data should only be used with caution. Further exploration/research into sample extraction and cleanup methods is necessary to derive useful pesticide quantification information from *Corbicula fluminea* samples. Pesticide data collected in this study are not considered usable.

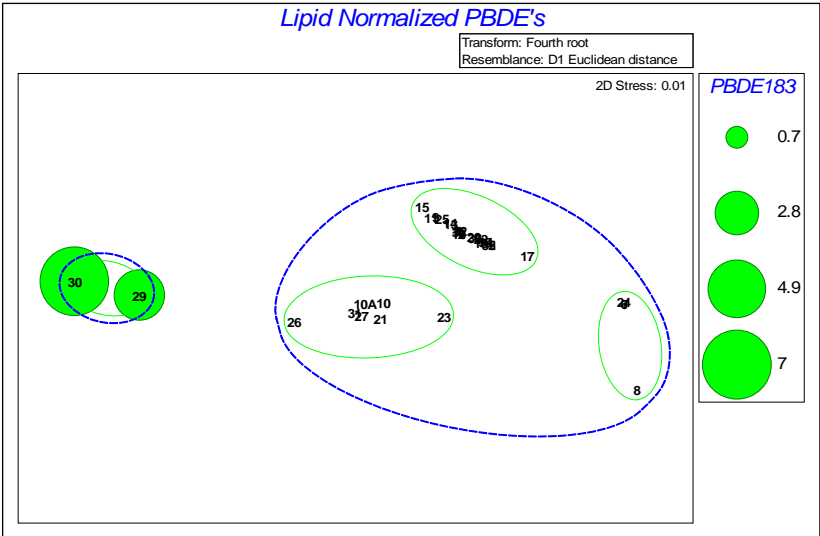
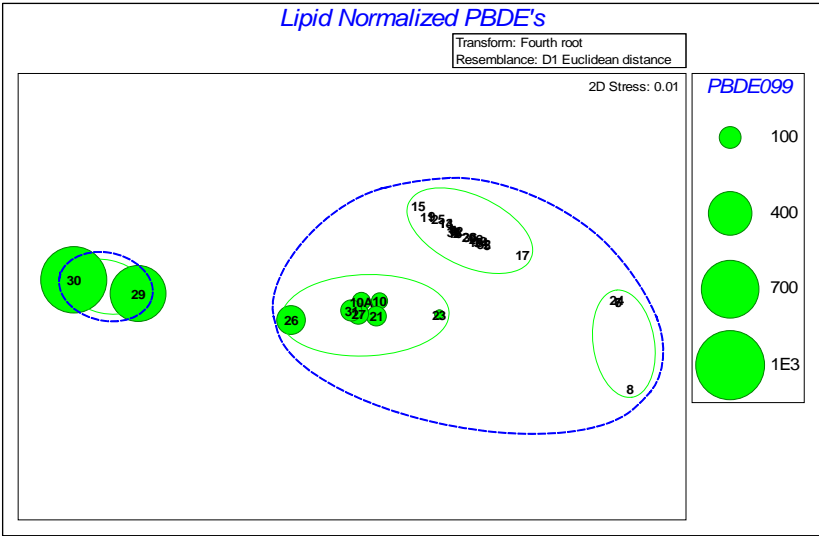
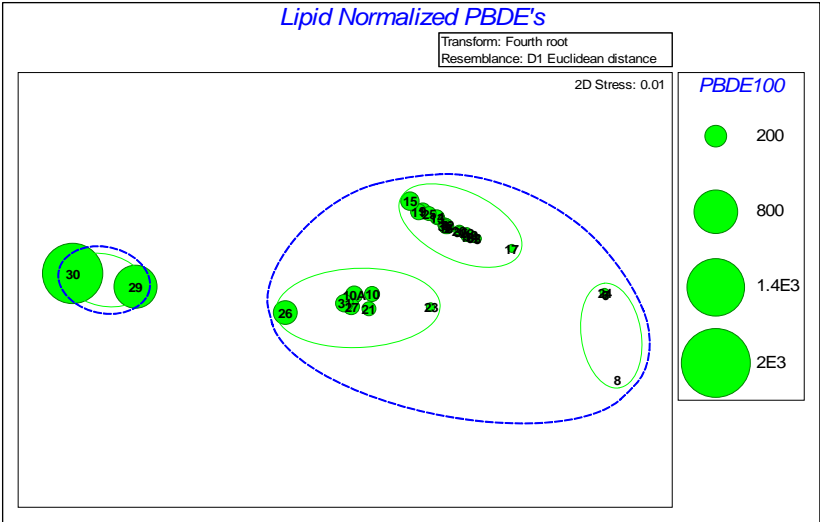
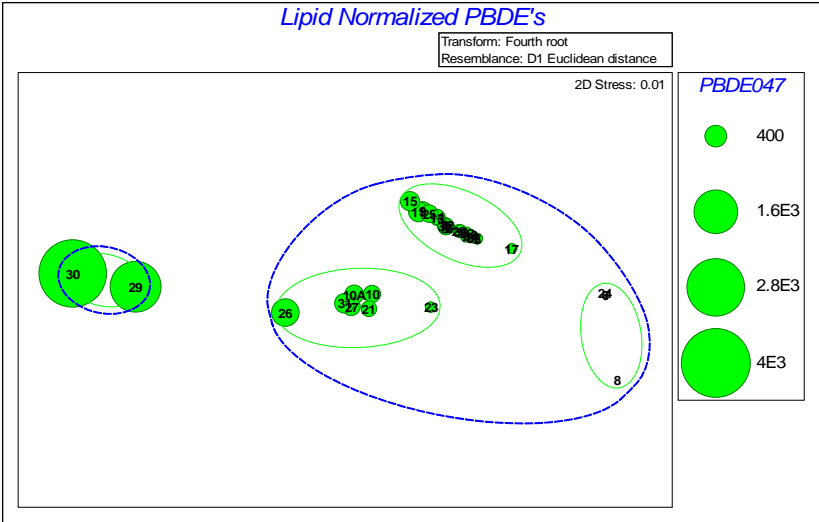
Appendix C: MDS Plots

Statistical analysis of individual analytes as well as total analytes as described in the report was conducted using Multidimensional Scaling (MDS). As part of the MDS approach, two-dimensional plots or biplots can be drawn to visualize the clustering and grouping of discrete samples and sample locations. The purpose of this analysis is to show the relative similarity of different sample locations in the composition and magnitude of analytes detected in the tissues of *Corbicula fluminea*. The biplots shown here were selected from a comprehensive set and represent most of the different spatial patterns observed for the relevant analytes.









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13. SUPPLEMENTARY NOTES					
14. ABSTRACT The freshwater Asiatic Clam, <i>Corbicula fluminea</i> , is found in abundance throughout North America. <i>C. fluminea</i> are primarily considered filter-feeders; however, they routinely bury in the sediment for extended periods and filter interstitial sediment water (pore water) or pedal-feed. <i>C. fluminea</i> shows promise as a model trophic-niche freshwater test organism or as an indicator species for bioaccumulation studies for the assessment of contaminants in sediments as part of dredging, restoration, remediation, and monitoring evaluations. In August and September 2005, 32 nearshore locations were sampled for <i>C. fluminea</i> along the Columbia River from Vista Park near Skamokawa, Washington (River Mile 32) to Warrendale, Oregon (River Mile 147). Four additional samples were collected in the lower Willamette River, near its confluence with the Columbia River (Columbia River Mile 102). Tissue samples were analyzed for semi-volatile compounds (including polycyclic aromatic hydrocarbons, PAH); chlorinated pesticides; polychlorinated biphenyl (PCB Aroclors and 209 congeners); polybrominated diphenyl ethers (PBDE; fire retardants); organotins; and four metals (Hg, Pb, Zn, Cd). All clam tissue had detectable levels of many of the chemicals analyzed. Statistical relationships among sampling stations were elucidated using exploratory multivariate statistical techniques. Relative abundances of major constituents were superimposed on regional maps displaying the sampling stations. A mid-reach point source for PCBs was identified, as were localized areas of DDTs, PBDEs, and PAHs.					
15. SUBJECT TERMS Bioaccumulation <i>Corbicula fluminea</i>		Columbia River Contaminants Sediment contamination			
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